# WEST

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**Search Results** - Record(s) 1 through 12 of 12 returned.

☐ 1. Document ID: US 5958718 A

L2: Entry 1 of 12 File: USPT Sep 28, 1999

US-PAT-NO: 5958718
DOCUMENT-IDENTIFIER: US 5958718 A
TITLE: Diagnosis and treatment of neuro-cognitive disorders associated with systemic immunological malfunction

Full | Title | Citation | Front | Review | Classification | Date | Reference | Claims | KWIC | Draw Desc | Image

☐ 2. Document ID: US 5593973 A

L2: Entry 2 of 12 File: USPT Jan 14, 1997

US-PAT-NO: 5593973
DOCUMENT-IDENTIFIER: US 5593973 A
TITLE: Treatment of viral hepatitis with mismatched dsRNA

Full | Title | Citation | Front | Review | Classification | Date | Reference | Claims | KWIC | Draw Desc | Image

☐ 3. Document ID: US 5258369 A

L2: Entry 3 of 12 File: USPT Nov 2, 1993

US-PAT-NO: 5258369
DOCUMENT-IDENTIFIER: US 5258369 A
TITLE: Treatment of chronic cerebral dysfunction by dsRNA methodology

Full | Title | Citation | Front | Review | Classification | Date | Reference | Claims | KWIC | Draw Desc | Image

☐ 4. Document ID: US 5194245 A

L2: Entry 4 of 12 File: USPT Mar 16, 1993

US-PAT-NO: 5194245
DOCUMENT-IDENTIFIER: US 5194245 A
TITLE: Diagnosis of viral hepatitis

Full | Title | Citation | Front | Review | Classification | Date | Reference | Claims | KWIC | Draw Desc | Image

☐ 5. Document ID: US 5132292 A

L2: Entry 5 of 12 File: USPT Jul 21, 1992

US-PAT-NO: 5132292
DOCUMENT-IDENTIFIER: US 5132292 A
TITLE: Treatment of viral hepatitis

Full | Title | Citation | Front | Review | Classification | Date | Reference | Claims | KWIC | Draw Desc | Image

☐ 6. Document ID: US 5063209 A

L2: Entry 6 of 12 File: USPT Nov 5, 1991

US-PAT-NO: 5063209
DOCUMENT-IDENTIFIER: US 5063209 A
TITLE: Modulation of aids virus-related events by double-stranded RNAs

Full | Title | Citation | Front | Review | Classification | Date | Reference | Claims | KWIC | Draw Desc | Image

☐ 7. Document ID: US 4963532 A

L2: Entry 7 of 12 File: USPT Oct 16, 1990

US-PAT-NO: 4963532
DOCUMENT-IDENTIFIER: US 4963532 A
TITLE: dsRNA-based prevention of viral escape

Full | Title | Citation | Front | Review | Classification | Date | Reference | Claims | KWIC | Draw Desc | Image

☐ 8. Document ID: US 4820696 A

L2: Entry 8 of 12 File: USPT Apr 11, 1989

US-PAT-NO: 4820696
DOCUMENT-IDENTIFIER: US 4820696 A
TITLE: Modulation of aids virus-related events by double-stranded RNAS

Full | Title | Citation | Front | Review | Classification | Date | Reference | Claims | KWIC | Draw Desc | Image

☐ 9. Document ID: US 4795744 A

L2: Entry 9 of 12 File: USPT Jan 3, 1989

US-PAT-NO: 4795744
DOCUMENT-IDENTIFIER: US 4795744 A
TITLE: Modulation of AIDS virus-related events by double-stranded RNAS

Full | Title | Citation | Front | Review | Classification | Date | Reference | Claims | KWIC | Draw Desc | Image

☐ 10. Document ID: EP 318281 A, AU 8825183 A, CA 1336684 C, JP 02111723 A, US 4963532 A, ZA 8808732 A

L2: Entry 10 of 12 File: DWPI May 31, 1989

DERWENT-ACC-NO: 1989-159331
DERWENT-WEEK: 198922
COPYRIGHT 2001 DERWENT INFORMATION LTD

TITLE: DS RNA used for prevention of viral escape - esp. in human immuno deficiency virus infection, by preventing the virus altering its host range or susceptibility to therapy

Full | Title | Citation | Front | Review | Classification | Date | Reference | Claims | KWIC | Draw Desc | Image

☐ 11. Document ID: AU 724056 B, EP 306347 A, NO 8803868 A, AU 8821864 A, DK 8804910 A, FI 8804069 A, HU 48029 T, ZA 8806581 A, JP 01131118 A, PT 88415 A, PT 91094 A, CN 1031651 A, DK 8903322 A, AU 8937811 A, CN 1039722 A, ZA 8905143 A, ES 2018903 A, AU 9217366 A, RU 2001917 C1, IL 90875 A, AU 9468836 A, IL 87664 A, AU 9510014 A, EP 306347 B1, DE 3853755 G, IE 63927 B, CA 1336683 C, CA 1336685 C, PH 26320 A, JP 96025884 B2, IE 68229 B, US 5593973 A, AU 684288 B, AU 9748499 A, NZ 226033 A

L2: Entry 11 of 12 File: DWPI Sep 14, 2000

DERWENT-ACC-NO: 1989-070509
DERWENT-WEEK: 200051
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TITLE: Diagnosis of double-stranded RNA deficiency states - using mismatched double-stranded RAN, opt. administered with an interferon e.g. to treat hepatitis

Full | Title | Citation | Front | Review | Classification | Date | Reference | Claims | KWIC | Draw Desc | Image

☐ 12. Document ID: EP 213921 A, JP 62077334 A, DK 8604052 A, ZA 8606418 A, CN 8605436 A, EP 213921 B, DE 3673288 G, CA 1326450 C, JP 95017510 B2, DK 170139 B

L2: Entry 12 of 12 File: DWPI Mar 11, 1987

DERWENT-ACC-NO: 1987-066579
DERWENT-WEEK: 199739
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TITLE: Double-stranded RNA against retrovirus and t-cell lymphotropic virus - useful for restoring suppressed immune state in aids

Full | Title | Citation | Front | Review | Classification | Date | Reference | Claims | KWIC | Draw Desc | Image

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| CARTER.DWPI,EPAB,JPAB,UG71,UG72,UG73,UG74,UG75,UG76,UG77,UG78,UG79,UG80,UG81 |
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| CARTERCOPTERS.DWPI,EPAB,JPAB,UG71,UG72,UG73,UG74,UG75,UG76,UG77,UG78,UG79,U |
| (CARTER$.IN. AND DSRNA AND THERAPY AND DEFICIENCY ).USPT,JPAB,EPAB,DWPI. |

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=> s rna and antisense

L1 26433 RNA AND ANTISENSE

=> s dsRNA or (double(s) stranded(s) RNA)

L2 26594 DSRNA OR (DOUBLE(S) STRANDED(S) RNA)

=> s l2 and antisense

L3 698 L2 AND ANTISENSE

=> s l3 and triplex

L4 37 L3 AND TRIPLEX

=> dup rem l4

PROCESSING COMPLETED FOR L4
L5 18 DUP REM L4 (19 DUPLICATES REMOVED)

=> d l5 ibib abs tot

L5 ANSWER 1 OF 18 MEDLINE DUPLICATE 1
ACCESSION NUMBER: 2000206927 MEDLINE
DOCUMENT NUMBER: 20206927
TITLE: Studies on anti-human immunodeficiency virus oligonucleotides that have alternating methylphosphonate/phosphodiester linkages.
AUTHOR: Miller P S; Cassidy R A; Hamma T; Kondo N S
CORPORATE SOURCE: Department of Biochemistry and Molecular Biology, School of
Hygiene and Public Health, Johns Hopkins University, 615

North Wolfe Street, Baltimore, MD, USA.. pmiller@jhsph.edu
CONTRACT NUMBER: GM57140 (NIGMS)
GM00664 (NIGMS)
SOURCE: PHARMACOLOGY AND THERAPEUTICS, (2000 Mar) 85 (3) 159-63.
Ref: 21
Journal code: P44. ISSN: 0163-7258.
PUB. COUNTRY: ENGLAND: United Kingdom
Journal; Article; (JOURNAL ARTICLE)
General Review; (REVIEW)
(REVIEW, TUTORIAL)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200009
ENTRY WEEK: 20000901

AB Preliminary investigations of the physical properties of oligonucleotide analogs that contain alternating methylphosphonate/phosphodiester linkages are described. An alternating oligo-2'-O-methylribonucleoside methylphosphonate, oligomer 1676, whose sequence is complementary to the upper hairpin region of human immunodeficiency virus TAR **RNA**, has been synthesized. This 15-mer forms a very stable duplex with its complementary **RNA** target, whose melting temperature is 71 degrees C. Introduction of two mismatched bases reduces the melting temperature by 16 degrees C. Similar results were obtained with the all-phosphodiester version of oligomer 1676, which demonstrates that introduction of the methylphosphonate linkages does not significantly perturb the ability of the oligo-2'-O-methylribonucleoside methylphosphonate to bind to **RNA**. Unlike the phosphodiester oligomer, however, oligomer 1676 is completely resistant to hydrolysis by the 3'-exonuclease activity found in mammalian serum. The interactions between nuclease-resistant, 5'-psoralen-derivatized, alternating oligo-2'-deoxypyrimidine methylphosphonates and **double-stranded** DNA were also studied. A 15-mer that contains thymine, 5-methylcytosine, and 5-propynyl-uracil forms a **triplex** with a polypurine tract found in the env gene of human immunodeficiency virus proviral DNA with an apparent dissociation constant of 400 nM at 22 degrees C. Maximal **triplex** formation by these oligomers is observed at approximately 2.5 mM magnesium, whereas maximal **triplex** formation by the corresponding all-phosphodiester oligomers occurs between 10 and 20 mM magnesium. This reduced magnesium dependence most likely results from reduced charge repulsion between the backbones of the methylphosphonate oligomer and purine strand of the target. The nuclease stability and ability of the methylphosphonate oligomers to form stable complexes with their target nucleic acids suggest that these oligomers are potential candidates for use as **antisense** or antigene agents in cell culture.

L5 ANSWER 2 OF 18 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 2000:293236 CAPLUS
TITLE: Sequence specific recognition of DNA by a .beta.-aminoalanine modified nucleic acid analog
AUTHOR(S): Fujii, Masayuki
CORPORATE SOURCE: Dept. of Applied Chemistry, Kinki University, Japan
SOURCE: Kinki Daigaku Kyushu Kogakubu Kenkyu Hokoku, Rikogaku-hen (2000), 28, 99-104
CODEN: KDKREY; ISSN: 0288-738X
PUBLISHER: Kinki Daigaku Kyushu Kogakubu
DOCUMENT TYPE: Journal
LANGUAGE: English

AB As substitutes for **antisense** and **triplex** oligonucleotides, oligopeptides contg. N.beta.-(thymin-1-ylacetyl)-.beta.-aminoalanine and n.beta.-(cytosin-1-ylacetyl)-.beta.-aminoalanine moieties were synthesized by solid phase synthesis using std. Boc chem. Obtained

20 mer peptide and 30 mer peptide, contg. 1-T and 10T/5C bases resp., showed hybridization properties with single **stranded** DNA and **RNA**, and also with **double stranded** DNA, at pH 7.0.

REFERENCE COUNT: 9
REFERENCE(S):
(1) Almarsson, O; Proc Natl Acad Sci USA 1993, V90, P9542 CAPLUS
(3) Dueholm, K; J Org Chem 1994, V59, P5767 CAPLUS
(5) Helene, C; Biochimica et Biophysica Acta 1990, V1049, P99 CAPLUS
(6) Hyrup, B; J Am Chem Soc 1994, V116, P7964 CAPLUS
(7) Moser, H; Science 1987, V238, P645 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L5 ANSWER 3 OF 18 MEDLINE DUPLICATE 2
ACCESSION NUMBER: 97388552 MEDLINE
DOCUMENT NUMBER: 97388552
TITLE: 2',5'-linked oligo-3'-deoxyribonucleoside phosphorothioate chimeras: thermal stability and **antisense** inhibition of gene expression.
AUTHOR: Bhan P; Bhan A; Hong M; Hartwell J G; Saunders J M; Hoke G D
CORPORATE SOURCE: Dyad Pharmaceutical Corporation, 9110 Red Branch Road, Columbia, MD 21045, USA.. purshotam.bhan@am.pharmacia.com
CONTRACT NUMBER: R44 GM49581-02 (NIGMS)
SOURCE: NUCLEIC ACIDS RESEARCH, (1997 Aug 15) 25 (16) 3310-7. Journal code: O8L. ISSN: 0305-1048.
PUB. COUNTRY: ENGLAND: United Kingdom
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Cancer Journals
ENTRY MONTH: 199711
ENTRY WEEK: 19971103
AB 2',5'-Linked oligo-3'-deoxyribonucleotides bind selectively to complementary **RNA** but not to DNA. These oligonucleotides (ODNs) do not recognize **double-stranded** DNA by Hoogsteen **triplex** formation and the complexes formed by these ODNs with **RNA** are not substrates for Escherichia coli RNase H. Substitution of the 2',5'-phosphodiester backbone by phosphorothioate linkages gives 2',5'-linked oligo-3'-deoxynucleoside phosphorothioate ODNs that exhibit significantly less non-specific binding to cellular proteins or thrombin. Incorporation of a stretch of seven contiguous 3',5'-linked oligo-2'-deoxynucleoside phosphorothioate linkages in the center of 2',5'-linked ODNs (as a putative RNase H recognition site) afford chimeric **antisense** ODNs that retain the ability to inhibit steroid 5alpha-reductase (5alphaR) expression in cell culture.

L5 ANSWER 4 OF 18 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1997:444917 CAPLUS
DOCUMENT NUMBER: 127:186216
TITLE: Modulation of nucleic acid structure by ligand binding: induction of a DNA.cntdot.RNA.cntdot.DNA hybrid **triplex** by DAPI intercalation
AUTHOR(S): Xu, Zhitao; Pilch, Daniel S.; Srinivasan, A. R.; Olson, Wilma K.; Geacintov, Nicholas E.; Breslauer, Kenneth J.
CORPORATE SOURCE: Department of Chemistry, Rutgers-The State University of New Jersey, New Brunswick, NJ, 08903, USA
SOURCE: Bioorg. Med. Chem. (1997), 5(6), 1137-1147
CODEN: BMECEP; ISSN: 0968-0896
PUBLISHER: Elsevier
DOCUMENT TYPE: Journal
LANGUAGE: English
AB The arom. diamidine, DAPI (4',6-diamidino-2-phenylindole), is used as an important biol. and cytol. tool since it forms highly fluorescent

complexes with nucleic acid duplexes via minor groove-directed/intercalative modes of interaction. In this study, we find that DAPI binding can induce the formation of an RNA-DNA hybrid **triplex** that would not otherwise form. More specifically, through application of a broad range of spectroscopic, viscometric, and mol. modeling techniques, we demonstrate that DAPI intercalation induces the formation of the poly(dT).cntdot.poly(rA).cntdot.poly(dT) hybrid triple helix, a structure which does not form in the absence of the ligand. Using UV mixing studies, we demonstrate that, in the presence of DAPI, the poly(rA).cntdot.poly(dT) duplex and the poly(dT) single strand form a 1:1 complex (a **triplex**) that does not form in the absence of DAPI. Through temp.-dependent absorbance measurements, we show that the poly(dT).cntdot.poly(rA).cntdot.poly(dT) **triplex** melts via two distinct transitions: initial conversion of the **triplex** to the duplex state, with the DAPI remaining bound, followed by denaturation of the duplex-DAPI complex to its component single strands and free DAPI. Using optical melting profiles, we show that DAPI binding enhances the thermal stability of the poly(dT).cntdot.poly(rA).cntdot.poly(dT) **triplex**, an observation consistent with the preferential binding of the ligand to the **triplex** vs. the duplex and single-stranded states. Our differential scanning calorimetric measurements reveal melting of the DAPI-satd. poly(dT).cntdot.poly(rA).cntdot.poly(dT) **triplex** to be assocd. with a lower enthalpy but greater cooperativity than melting of the corresponding DAPI-satd. poly(rA).cntdot.poly(dT) duplex. Our flow linear dichroism and viscometric data are consistent with an intercalative mode of binding when DAPI interacts with both the poly(dT).cntdot.poly(rA).cntdot.poly(dT) **triplex** and the poly(rA).cntdot.poly(dT) duplex. Finally, computer modeling studies suggest that a combination of both stacking and electrostatic interactions between the intercalated ligand and the host nucleic acid play important roles in the DAPI-induced stabilization of the poly(dT).cntdot.poly(rA).cntdot.poly(dT) **triplex**. In the aggregate, our results demonstrate that ligand binding can be used to induce the formation of **triplex** structures that do not form in the absence of the ligand. This **triplex**-inducing capacity has potentially important implications in the design of novel **antisense**, antigene, antiviral, and diagnostic strategies.

L5 ANSWER 5 OF 18 MEDLINE

| | |
|---|---|
| ACCESSION NUMBER: | 1998247171 MEDLINE |
| DOCUMENT NUMBER: | 98247171 |
| TITLE: | Inhibition of HIV-1 replication by foldback triple-helix forming oligonucleotides. |
| AUTHOR: | Hiratou T; Tsukahara S; Takai K; Koyanagi Y; Yamamoto N; Takaku H |
| CORPORATE SOURCE: | Department of Industrial Chemistry, Chiba Institute of Technology, Japan. |
| SOURCE: | NUCLEIC ACIDS SYMPOSIUM SERIES, (1997) (37) 221-2. Journal code: O8N. ISSN: 0261-3166. |
| PUB. COUNTRY: | ENGLAND: United Kingdom Journal; Article; (JOURNAL ARTICLE) |
| LANGUAGE: | English |
| FILE SEGMENT: | Priority Journals |
| ENTRY MONTH: | 199809 |
| ENTRY WEEK: | 19980904 |

AB Replication of retroviral **RNA** into **double-stranded** DNA is catalyzed by reverse transcriptase (RT). The polypurine tract (PPT) serves as a primer for plus-strand DNA synthesis and is highly conserved among HIV-1. The PPT region is a possible target for triple-helix formation. Here, we show the effects of triple-helix formation by analyses of melting temperature and gel shift using a foldback **triplex**-forming-oligonucleotides (FTFOs). We found that the FTFOs containing phosphorothioate groups at the 3'- and 5'-ends, or

inside the hairpin loop, exhibited greater exonuclease resistance than the unmodified FTFOs. Several **triplex** oligonucleotides have thermal stability. The abilities of the FTFOs (DsDG-37) containing the guanosine in place of the cytidine in the third Hoogsteen base-pairing strand to inhibit HIV-1 replications were examined. The FTFOs (DsDG-37) inhibit the replication of HIV-1 more efficiently than the FTFOs (DsD-37) indicating sequence-specific inhibition of HIV-1 replication.

L5 ANSWER 6 OF 18 MEDLINE DUPLICATE 3
ACCESSION NUMBER: 96394554 MEDLINE
DOCUMENT NUMBER: 96394554
TITLE: Double hairpin complexes allow accommodation of all four base pairs in triple helices containing both DNA and RNA strands.
AUTHOR: Pascolo E; Toulme J J
CORPORATE SOURCE: INSERM U.386, IFR Pathologies Infectieuses, Universite Victor Segalen Bordeaux II, 146 rue Leo Saignat, 33076 Bordeaux cedex, France.
SOURCE: JOURNAL OF BIOLOGICAL CHEMISTRY, (1996 Sep 27) 271 (39) 24187-92.
Journal code: HIV. ISSN: 0021-9258.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Cancer Journals
ENTRY MONTH: 199701
ENTRY WEEK: 19970104
AB We investigated the binding of an **antisense** oligodeoxynucleotide to a stem-loop structure corresponding to the mini-exon sequence of the protozoan parasite Leishmania amazonensis. This oligomer was designed to anneal to the single-**stranded** region adjacent to the bottom of the hairpin and to fold back on itself, giving rise to a "**double** -hairpin" complex that involved a local **triplex**. This imposed the recognition, by the third strand, of a "purine" strand containing 6 interspersed pyrimidines out of 15 nucleic acid bases. The sequence of the complementary oligonucleotide was derived from the so-called pyrimidine motif; the third strand of the anti-mini-exon oligomer was parallel to the purine strand of the target. Electrophoretic mobility shift assays and footprinting studies demonstrated that such an **antisense** oligomer was able to bind to both the DNA and **RNA** versions of the Leishmania hairpin. These **double** hairpin complexes allowed the formation at pH 6.0 of a triple-**stranded** structure, despite the presence of 4 A:T*G and 2 G:C*T triplets out of 15.

L5 ANSWER 7 OF 18 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1997:269065 CAPLUS
DOCUMENT NUMBER: 126:313724
TITLE: Hybridization properties of nucleic acid analogs bearing peptide backbone
AUTHOR(S): Fujii, Masayuki; Yoshida, Kohya; Hidaka, Jinsai; Ohtsu, Takayuki
CORPORATE SOURCE: Department of Industrial Chemitsry, Faculty of Engineering in Kyushu, Kinki University, Iizuka, 820, Japan
SOURCE: Pept. Chem. (1996), 34th, 461-464
CODEN: PECHDP; ISSN: 0388-3698
PUBLISHER: Protein Research Foundation
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Two types of nucleic acid analog peptides (NAP), as substitutes for **antisense** and **triplex** oligonucleotides were prepd. by solid phase synthesis using std. Boc chem., and their hybridization properties were investigated by means of Tm measurement. UV melting

curves revealed that they showed hybridization properties with single **stranded** DNA and **RNA**, and also with **double stranded** DNA.

L5 ANSWER 8 OF 18 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1996:683903 CAPLUS
DOCUMENT NUMBER: 126:436
TITLE: Inhibition of HIV-1 replication by oligonucleotides forming triple-helixes targeted to polypurine tract
AUTHOR(S): Tsukahara, Satoru; Suzuki, Junji; Goto, Yuta; Inagawa, Takubumi; Takeuchi, Hiroaki; Takai, Kazuyuki; Koyanagi, Yoshio; Yamamoto, Naoki; Takaku, Hiroshi
CORPORATE SOURCE: Dep. Industrial Chem., Chiba Inst. Technol., Chiba, 275, Peop. Rep. China
SOURCE: Nucleic Acids Symp. Ser. (1996), 35(Twentythird Symposium on Nucleic Acids Chemistry, 1996), 181-182
CODEN: NACSD8; ISSN: 0261-3166
PUBLISHER: Oxford University Press
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Replication of retroviral **RNA** into **double-stranded** DNA is catalyzed by reverse transcriptase (RT). The polypurine tract (PPT) serves as a primer for plus-strand DNA synthesis and is highly conserved among HIV-1. The PPT region is a possible target for triple-helix formation. Here, we show the effects of triple-helix formation by analyses of melting temp. and protection from reverse transcription in vitro using two systems (two-strand or three-strand-system). Furthermore, we used phosphorothioate oligonucleotide probes to increase the nuclease resistance. Several **triplex** oligonucleotides have thermal stability and prevent the initiation of minus-strand DNA synthesis by RT. We also demonstrate inhibition of HIV-1 replication by these oligonucleotides.

L5 ANSWER 9 OF 18 MEDLINE DUPLICATE 4
ACCESSION NUMBER: 95249361 MEDLINE
DOCUMENT NUMBER: 95249361
TITLE: Single strand targeted **triplex**-formation. Destabilization of guanine quadruplex structures by foldback **triplex**-forming oligonucleotides.
AUTHOR: Kandimalla E R; Agrawal S
CORPORATE SOURCE: Hybridon, Inc., Worcester, MA 01605, USA.
SOURCE: NUCLEIC ACIDS RESEARCH, (1995 Mar 25) 23 (6) 1068-74.
Journal code: O8L. ISSN: 0305-1048.
PUB. COUNTRY: ENGLAND: United Kingdom
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Cancer Journals
ENTRY MONTH: 199508
AB Oligonucleotides that can hybridize to single-**stranded** complementary polypurine nucleic acid targets by Watson-Crick base pairing as well as by Hoogsteen base pairing, referred to here as foldback **triplex**-forming oligonucleotides (FTFOs), have been designed. These oligonucleotides hybridize with target nucleic acid sequences with greater affinity than **antisense** oligonucleotides, which hybridize to the target sequence only by Watson-Crick hydrogen bonding [Kandimalla, E. R. and Agrawal, S. Gene(1994) 149, 115-121 and references cited therein]. FTFOs have been studied for their ability to destabilize quadruplexes formation by **RNA** or DNA target sequences. The influence of various DNA/**RNA** compositions of FTFOs on their ability to destabilize **RNA** and DNA quadruplexes has been examined. The ability of the FTFOs to destabilize quadruplex structures is related to the structurally and thermodynamically stable foldback **triplex** formed between the FTFO and its target sequence.

**Antisense** oligonucleotides (DNA or **RNA**) that can form only a Watson-Crick **double** helix with the target sequence are unable to destabilize quadruplex structures of **RNA** and DNA target sequences and are therefore limited in their repertoire of target sequences. The quadruplex destabilization ability of FTFOs is dependent on the nature of the cation present in solution. The **RNA** quadruplex destabilization ability of FTFOs is -20% higher in the presence of sodium ion than potassium ion. The use of FTFOs, which can destabilize quadruplex structure, opens up new areas for development of oligonucleotide-based therapeutics, specifically, targeting guanine-rich sequences that exist at the ends of pro- and eukaryotic chromosomes and dimerization regions of retroviral **RNA**.

L5 ANSWER 10 OF 18 MEDLINE DUPLICATE 5
ACCESSION NUMBER: 94359804 MEDLINE
DOCUMENT NUMBER: 94359804
TITLE: Pyrimidine phosphorothioate oligonucleotides form triple-stranded helices and promote transcription inhibition.
AUTHOR: Xodo L; Alunni-Fabbroni M; Manzini G; Quadrifoglio F
CORPORATE SOURCE: Department of Biochemistry, Biophysics and Macromolecular Chemistry, University of Trieste, Italy..
SOURCE: NUCLEIC ACIDS RESEARCH, (1994 Aug 25) 22 (16) 3322-30. Journal code: O8L. ISSN: 0305-1048.
PUB. COUNTRY: ENGLAND: United Kingdom
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Cancer Journals
ENTRY MONTH: 199412

AB The ability of phosphorothioate (POS) oligonucleotides to recognise and bind to homopurine-homopyrimidine DNA **double-stranded** sites via triple helix formation has been investigated. It has been found that the homologous pyrimidine POS sequences Y11-Si (i = 0, 1,2,3,4,10), which have been obtained by an increasing sulphur substitution in the sugar-phosphate backbone of d(CTTCCTCCTCT) (Y11), and the target hairpin duplex d(GAAGGAGGAGA-T4-TCTCCTCCTTC) (h26) can form stable triple helices, as indicated by PAGE, CD and UV melting experiments. The thermal stability of the triple helices depends on the number of POS linkages in the third Y11 strand, varying from 48 degrees C (Y11, with only phosphate groups, PO2) to 31 degrees C (Y11-S10 containing exclusively thioate groups). On average, a Tm depression of about 2 degrees C per POS linkage introduced in Y11 was observed. CD data indicate that the sulphurization of the third strand results in minimal changes of triple-**stranded** structures. The energetics of the **triplex**-to-hairpin plus single-strand transition has been determined by van't Hoff analyses of the melting curves. In free energy terms, the POS triplexes h26.Y11-Si are less stable than the normal PO2 h26.Y11 **triplex** by values between 2.7 and 5.4 kcal/mol, depending on the number of POS linkages contained in the third strand. Phosphorothioate oligonucleotides being resistant towards several nucleases offer an interesting choice as gene blockers in **antisense** strategy. Thus, their ability to inhibit transcription via triple helix formation has been examined in vitro. We found that **triplex**-forming POS oligonucleotides of 20 bases in length (with a cytosine contents of 45%), containing either 10% or 26% thioate groups, strongly repress the transcription activity of the bacteriophage T7 **RNA** polymerase at pH 6.9, when used in excess compared to the target (mol oligo/mol template = 125). The here reported data are useful for designing phosphorothioate oligonucleotides targeted to genomic DNA in

antigene strategy.

L5 ANSWER 11 OF 18 MEDLINE DUPLICATE 6
ACCESSION NUMBER: 95047456 MEDLINE
DOCUMENT NUMBER: 95047456
TITLE: Single-strand-targeted **triplex** formation: stability, specificity and RNase H activation properties.
AUTHOR: Kandimalla E R; Agrawal S
CORPORATE SOURCE: Hybridon, Inc., Worcester, MA 01605..
SOURCE: GENE, (1994 Nov 4) 149 (1) 115-21.
Journal code: FOP. ISSN: 0378-1119.
PUB. COUNTRY: Netherlands
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199502

AB Single-**stranded** (ss) oligodeoxyribonucleotides (oligos) containing both Watson-Crick and Hoogsteen hydrogen bonding domains joined by either a 5-nucleotide loop or a flexible hexaethylene-glycol linker, called foldback **triplex**-forming oligos (FTFOs), are designed and studied for their binding affinity and specificity to their ss DNA/**RNA** targets. Thermal denaturation studies revealed an increased affinity of FTFOs, due to addition of a Hoogsteen hydrogen bonding domain at the binding site, as the Watson-Crick domain forms a **double** helix with the target, when compared to conventional **antisense** and antigene oligos. DNase I hydrolysis and electrophoretic mobility shift analysis confirmed the formation of foldback triplexes relative to conventional **double**- and triple-**stranded** structures. The FTFOs showed increased sequence specificity mainly arising from their ability to recognize the target sequence twice, first by Watson-Crick base pairing and a second time by Hoogsteen base pairing. An FTFO with DNA components in both duplex- and **triplex**-forming domains showed preference for a DNA homopurine target strand.

L5 ANSWER 12 OF 18 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1994:153703 CAPLUS
DOCUMENT NUMBER: 120:153703
TITLE: Treatment of cellular hyperproliferation by inhibition of interleukin-1
INVENTOR(S): Cooper, Kevin D.; Hammerberg, Craig; Maxwell, Kameron W.; Tseng, Ben Y.
PATENT ASSIGNEE(S): Genta Inc., USA; University of Michigan
SOURCE: PCT Int. Appl., 40 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|---|---|---|---|---|
| WO 9324134 | A1 | 19931209 | WO 1993-US4917 | 19930521 |
| W: AU, CA, JP, KR, NZ | | | | |
| RW: AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE | | | | |
| AU 9343889 | A1 | 19931230 | AU 1993-43889 | 19930521 |
| EP 644765 | A1 | 19950329 | EP 1993-914110 | 19930521 |
| R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LI, LU, MC, NL, PT, SE | | | | |
| JP 07508977 | T2 | 19951005 | JP 1993-500694 | 19930521 |
| PRIORITY APPLN. INFO.: | | | US 1992-887734 | 19920522 |
| | | | WO 1993-US4917 | 19930521 |

AB Benign or malignant pathol. hyperproliferation of skin or epithelial cells

is treated by exposing the cells to (a) an **antisense** oligomer complementary to **RNA** transcribed from a target gene in the cells, (b) a 3rd-strand oligomer complementary to a **double-stranded** target gene sequence, or (c) a **triplex** oligomer pair complementary to a single-**stranded** target gene sequence. The target gene encodes a cytokine mediating cellular proliferation, a cytokine-modulating factor, a cytokine-activating enzyme, or an enzyme involved in translational or posttranslational modification of the cytokine. Specifically, the target gene may encode interleukin-1.alpha. (IL-1.alpha.), IL-1.beta., intracellular IL-1 receptor antagonist, or an IL-1 converting enzyme. The oligomers may be used to treat psoriasis, inflammatory bowel or ocular diseases, or rheumatoid arthritis. Thus, proliferation of normal human keratinocytes was 69% inhibited by exposure for 7 days to 100 .mu.M TTCTGCCATGGCTGC methylphosphonate analog ( **antisense** oligomer for IL-1.beta. gene).

L5 ANSWER 13 OF 18 LIFESCI COPYRIGHT 2001 CSA
ACCESSION NUMBER: 95:12907 LIFESCI
TITLE: Crystal structure of a parallel-stranded duplex of a deoxycytidylyl-(3'-5')-deoxycytidine analogue containing intranucleosidyl C(3')-C(5') ethylene bridges
AUTHOR: Egli, M.; Lubini, P.; Bolli, M.; Dobler, M.; Leumann, C.
CORPORATE SOURCE: Organ. Chem. Lab., ETH Swiss Fed. Inst. Technol., ETH-Zentrum, CH-8092 Zuerich, Switzerland
SOURCE: J. AM. CHEM. SOC., (1993) vol. 115, no. 13, pp. 5855-5856.
ISSN: 0002-7863.
DOCUMENT TYPE: Journal
FILE SEGMENT: N
LANGUAGE: English
SUMMARY LANGUAGE: English

AB The proposal to use synthetic **antisense** oligonucleotides for therapeutic purposes has led to a great interest in the modification of natural DNA and **RNA** molecules by chemical methods. To probe the possibility of stabilizing duplex formation entropically by using **antisense** oligonucleotides with a conformationally more rigid sugar phosphate backbone as the complex partner for a natural DNA (or **RNA**) sequence, we designed and synthesized a new type of nucleosides (bicyclonucleosides) that differs from the natural deoxynucleosides by an additional ethylene bridge between the centers C(3') and C(5'). studies on homodecamers with the nucleobases adenine and thymine thereof essentially confirmed the expected (numerical) reduction of the entropy term upon duplex formation and furthermore revealed a higher propensity for **triplex** formation of these analogues. To obtain insight into the structural details of a bicyclo-DNA (bed) single strand, and thus into its preorganization for duplex formation, we synthesized and crystallized the corresponding dinucleotide analogue, bcd(C sub(2)), of deoxycytidylyl-(3'-5')-deoxycytidine and unexpectedly found it to form a parallel-**stranded**, right-handed duplex, paired via C-C super(+) base pairs with three hydrogen bonds. The cytosine base pairs are stacked at a distance of 3.44 angstrom with a helical twist of 34 degree . Beyond the scope of the investigation, this first high-resolution crystal structure of a homocytosine minihelix is reminiscent of the suggested molecular arrangement of **double-stranded** poly(dC) at neutral pH. It has been shown previously that poly(C) at low pH super(7) forms a parallel-oriented, base-paired duplex; however, no structural details are known so far. Parallel orientation of strands with G-G and C-C super(+) base pairs was previously observed in crystals of the duplex [d(CpG)] sub(2) grown at low pH, but the average distance of 4.34 angstrom between base pairs suggested only weak stacking interactions. Self-pairing of cytosine bases via three hydrogen bonds occurs in crystals of cytosine-5-acetic acid and cytosine hemitrichloroacetate.

L5 ANSWER 14 OF 18 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1993:626310 CAPLUS
DOCUMENT NUMBER: 119:226310
TITLE: Synthesis and binding properties of pyrimidine oligodeoxynucleoside analogs containing neutral phosphodiester replacements: the formacetal and 3'-thioformacetal internucleoside linkages
AUTHOR(S): Jones, Robert J.; Lin, Kuei Ying; Milligan, John F.; Wadwani, Shalini; Matteucci, Mark D.
CORPORATE SOURCE: Gilead Sci., Foster City, CA, 94404, USA
SOURCE: J. Org. Chem. (1993), 58(11), 2983-91
CODEN: JOCEAH; ISSN: 0022-3263
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Pyrimidine dimer deoxyribonucleosides contg. neutral phosphodiester with formacetal and 3'-thioformacetal internucleoside linkages, are prepd and incorporated into oligodeoxyribonucleosides (ODNs) . The binding properties ODNs to single-**stranded** (ss) **RNA** and **double-stranded** (ds) DNA were then detd. The triple helix formation properties of the 3'-thioformacetal and formacetal ODNs were detd. by footprint and restriction enzyme inhibition assays. The 3'-thioformacetal ODN binds to dsDNA with an affinity slightly less than the control ODN. The high affinity and specificity of an ODN contg. the 3'-thioformacetal for the ssRNA target and dsDNA target suggest that this linkage is a promising analog for both **antisense** and triple helix therapeutic applications.

L5 ANSWER 15 OF 18 LIFESCI COPYRIGHT 2001 CSA
ACCESSION NUMBER: 94:77261 LIFESCI
TITLE: A DNA hairpin as a target for **antisense** oligonucleotides
AUTHOR: Brossalina, E.; Toulme, J.-J.*
CORPORATE SOURCE: Lab. Biophys. Mol., INSERM CJF 90-13, Universite de Bordeaux II, 146 Rue Leo Saignat, 33076 Bordeaux Cedex, France
SOURCE: J. AM. CHEM. SOC., (1993) vol. 115, no. 2, pp. 796-797. ISSN: 0002-7863.
DOCUMENT TYPE: Journal
FILE SEGMENT: N; W3
LANGUAGE: English; English
AB Artificial regulation of gene expression can be achieved by **antisense** oligonucleotides complementary to part of a messenger **RNA**. Although RNAs can be written as single strands, self-pairing between adjacent or remote sequences gives rise to **double-stranded** regions. **RNA** hairpins will weaken or prevent the binding of an **antisense** oligomer if its target is sequestered in such a structure. We propose here a strategy to bind an oligonucleotide to a hairpin without disrupting the structure, via the formation of base triplets. Our suggestion is to form a "**double** hairpin" complex. The **antisense** oligomer has two domains: the first one is complementary to the single-**stranded** sequence at the bottom of the hairpin, and the second one is designed to form a **triplex** with both the hybridized first domain and the stem of the hairpin.

L5 ANSWER 16 OF 18 MEDLINE DUPLICATE 7
ACCESSION NUMBER: 93277979 MEDLINE
DOCUMENT NUMBER: 93277979
TITLE: The polypurine tract, PPT, of HIV as target for **antisense** and triple-helix-forming oligonucleotides.
AUTHOR: Volkmann S; Dannull J; Moelling K
CORPORATE SOURCE: Max-Planck-Institute fur Molekulare Genetik, Berlin, Germany.
SOURCE: BIOCHIMIE, (1993) 75 (1-2) 71-8.
Journal code: A14. ISSN: 0300-9084.

PUB. COUNTRY: France
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199309

AB Replication of retroviral **RNA** into **double-stranded** DNA provirus involves initiation of plus-strand DNA synthesis at the polypurine tract, PPT, by the reverse transcriptase (RT). The PPT is highly conserved among the known HIV-1 retroviral isolates. It occurs twice, once within the coding region of the integrase and the other one adjacent to the 3' LTR. The data presented show that two **antisense** oligonucleotides, a 20-mer and a 40-mer, complementary to the PPT induce complete blocks of DNA synthesis whereas an **antisense** oligonucleotide outside the PPT is only slightly inhibitory. Previously polypurine sequences have been used by several groups for **triplex**-formation. During replication the HIV-polypurine tract, PPT, is present in a **RNA**-DNA hybrid. Therefore triple-helix formation consisting of **RNA**-DNA and a third DNA strand covering the PPT region was tested here by protection against RNase H cleavage in vitro. Incubation with a pyrimidine oligonucleotide in parallel orientation to the PPT-**RNA** shows some protection. GT-pyrimidine-purine mixed oligonucleotides (25-mer) led to protection against RNase H up to 50% independent of their orientation. The data suggest that triple-helix formation may have taken place with the PPT in vitro. Therefore, this highly conserved structure may prove useful in nucleic acid based anti-viral therapy with **antisense** or triple-helix approaches. Furthermore, the influence of HIV-1 nucleocapsid (NC) protein, NCp15, on reverse transcription is reported. The data show a two- to three-fold stimulatory effect of the NCp15 on **RNA** directed DNA synthesis.

L5 ANSWER 17 OF 18 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1993:160365 CAPLUS
DOCUMENT NUMBER: 118:160365
TITLE: Nonionic oligonucleotide analogs (Matagen) as anticodic agents in duplex and **triplex** formation
AUTHOR(S): Ts'o, P. O. P.; Aurelian, L.; Chang, E.; Miller, P. S.
CORPORATE SOURCE: Sch. Hyg. Public Health, Johns Hopkins Univ., Baltimore, MD, 21205, USA
SOURCE: Ann. N. Y. Acad. Sci. (1992), 660(Antisense Strategies), 159-77
CODEN: ANYAA9; ISSN: 0077-8923
DOCUMENT TYPE: Journal; General Review
LANGUAGE: English

AB A review with 46 refs. The authors suggest that the biol. approach should retain the term "**antisense** approach," whereas the chem. approach should be termed the "anticode approach.". The targets of the anticode approach can be at either the single-**stranded RNA** level or the **double-stranded** DNA level. The anticode approach appears to be much more amendable to efficient development and rational design of therapeutic agents. The authors experiences over the last 20 yr in this area of anticode approach are briefly described.

L5 ANSWER 18 OF 18 MEDLINE DUPLICATE 8
ACCESSION NUMBER: 92126178 MEDLINE
DOCUMENT NUMBER: 92126178
TITLE: The anti-gene strategy: control of gene expression by **triplex**-forming-oligonucleotides.
AUTHOR: Hel`ene C

CORPORATE SOURCE: Laboratoire de Biophysique, Museum National d'Histoire Naturelle, INSERM U201-CNRS UA 481, Paris, France..
SOURCE: ANTI-CANCER DRUG DESIGN, (1991 Dec) 6 (6) 569-84. Ref: 60
Journal code: AC5. ISSN: 0266-9536.
PUB. COUNTRY: ENGLAND: United Kingdom
Journal; Article; (JOURNAL ARTICLE)
General Review; (REVIEW)
(REVIEW, TUTORIAL)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Cancer Journals
ENTRY MONTH: 199205

AB Oligonucleotides are being developed to selectively inhibit gene expression at the translational level (**antisense** oligonucleotides) and at the transcriptional level (anti-gene oligonucleotides). This review deals with the anti-gene strategy whereby an oligonucleotide binds to the major groove of **double** helical DNA where it forms a local triple helix. The molecular mechanisms for DNA recognition by triple helix formation are discussed together with some of the rules presently available to design the sequence and orientation of the triple helix forming oligonucleotide. **Triplex** stability can be enhanced by covalent attachment of an intercalating agent to the terminal nucleotide of the oligonucleotide. The intercalating agent can be used to induce irreversible reactions in the target sequence: **double** strand cleavage by a phenanthroline-Cu chelate in the presence of a reducing agent, photo-induced cleavage by ellipticine derivatives, photo-induced cross-linking of the two DNA strands by psoralen... Triple helix-forming oligonucleotides can be used to control gene expression at the transcriptional level. Inhibition of binding of transcription activating factors by **triplex** formation modulates the level of transcription of the target gene. Binding of a **triplex**-forming oligonucleotide immediately downstream of the **RNA** polymerase binding site can inhibit transcription initiation as shown with the E. coli beta-lactamase gene. Studies with cells in culture show that triple helix formation may occur in the intracellular environment and consequently leads to transcription inhibition. This inhibitory effect can be made irreversible by using, e.g., psoralen-substituted oligonucleotides. Oligonucleotides synthesized with the alpha-anomers of nucleotide units are resistant to nucleases and still form triple helices with **double-stranded** DNA. Oligo-[alpha]-deoxynucleotides can be derived by stabilizing (intercalating) agents or reactive groups (cleaving reagents, cross-linkers ...). The results presently available provide a rational basis for the development of new tools for cellular biology and of new therapeutical approaches to selectively control gene expression at the transcriptional level.

=> s cosuppression

L6 291 COSUPPRESSION

=> s l6 and elegans

L7 20 L6 AND ELEGANS

=> dup rem l7

PROCESSING COMPLETED FOR L7
L8 10 DUP REM L7 (10 DUPLICATES REMOVED)

=> d l8 ibib abs tot

L8 ANSWER 1 OF 10 MEDLINE DUPLICATE 1

ACCESSION NUMBER: 2000386785 MEDLINE
DOCUMENT NUMBER: 20347034
TITLE: Transgene-mediated **cosuppression** in the C. **elegans** germ line.
AUTHOR: Dernburg A F; Zalevsky J; Colaiacovo M P; Villeneuve A M
CORPORATE SOURCE: Departments of Developmental Biology and Genetics, Stanford
University School of Medicine, CA 94305-5329, USA.
SOURCE: GENES AND DEVELOPMENT, (2000 Jul 1) 14 (13) 1578-83.
Journal code: FN3. ISSN: 0890-9369.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200010
ENTRY WEEK: 20001002
AB Functional silencing of chromosomal loci can be induced by transgenes ( **cosuppression**) or by introduction of double-stranded RNA (RNAi). Here, we demonstrate the generality of and define rules for a transgene-mediated **cosuppression** phenomenon in the Caenorhabditis **elegans** germ line. Functional repression is not a consequence of persistent physical association between transgenes and endogenous genes or of mutations in affected genes. The **cosuppression** mechanism likely involves an RNA mediator that defines its target specificity, reminiscent of RNAi. **Cosuppression** is strongly abrogated in rde-2 and mut-7 mutants, but is not blocked in an
rde-1 mutant, indicating that **cosuppression** and RNAi have overlapping but distinct genetic requirements.

L8 ANSWER 2 OF 10 BIOSIS COPYRIGHT 2001 BIOSIS
ACCESSION NUMBER: 2000:346449 BIOSIS
DOCUMENT NUMBER: PREV200000346449
TITLE: Interfering with gene expression.
AUTHOR(S): Marx, Jean
SOURCE: Science (Washington D C), (26 May, 2000) Vol. 288, No. 5470, pp. 1370-1372. print.
ISSN: 0036-8075.
DOCUMENT TYPE: Article
LANGUAGE: English
SUMMARY LANGUAGE: English
AB An explosion of recent evidence is revealing a new cellular pathway for silencing specific genes at the messenger RNA level that may protect organisms against viruses and genetic damage.

L8 ANSWER 3 OF 10 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.DUPLICATE 2
ACCESSION NUMBER: 2000319653 EMBASE
TITLE: The silence of the genes.
AUTHOR: Plasterk R.H.A.; Ketting R.F.
CORPORATE SOURCE: R.H.A. Plasterk, Hubrecht Laboratory, Uppsalalaan 8, 3584 CT Utrecht, Netherlands. plasterk@niob.knaw.nl
SOURCE: Current Opinion in Genetics and Development, (2000) 10/5 (562-567).
Refs: 54
ISSN: 0959-437X CODEN: COGDET
COUNTRY: United Kingdom
DOCUMENT TYPE: Journal; General Review
FILE SEGMENT: 004 Microbiology
LANGUAGE: English
SUMMARY LANGUAGE: English
AB About two years ago, it was recognized that introduction of double-stranded RNA (dsRNA) had a potent effect on gene expression, in particular on mRNA stability. Since then, this process has been found to occur in many different organisms, and to bear a strong resemblance to a previously recognized process in plants, called **cosuppression**.
Both genetic and biochemical studies have started to unravel the mysteries

of RNA interference: genes involved in this process are being identified and in vitro studies are giving the first hints of what is happening to both the dsRNA and the affected mRNA molecules after the introduction of the dsRNA.

L8 ANSWER 4 OF 10 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 2000:199992 CAPLUS
DOCUMENT NUMBER: 133:71533
TITLE: Genetic analysis of RNA interference and transposon silencing in C. **elegans**
AUTHOR(S): Tabara, Hiroaki
CORPORATE SOURCE: Program Molecular Med., Univ. Massachusetts, Worcester, USA
SOURCE: Jikken Igaku (2000), 18(3), 360-362
CODEN: JIIGEF; ISSN: 0288-5514
PUBLISHER: Yodosha
DOCUMENT TYPE: Journal; General Review
LANGUAGE: Japanese
AB A review with 10 refs., on genetic anal. of the mechanism of RNA interference (RNAi); biol. role of RNAi; and relations between RNAi and **cosuppression** and quelling, with resp. to role of dsRNA in RNAi in Caenorhabditis elegan.

L8 ANSWER 5 OF 10 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 2000:222347 CAPLUS
DOCUMENT NUMBER: 132:319930
TITLE: A genetic link between co-suppression and RNA interference in C. **elegans**
AUTHOR(S): Ketting, Rene F.; Plaster, Ronald H. A.
CORPORATE SOURCE: Division of Molecular Biology, The Netherlands Cancer Institute, Centre for Biomedical Genetics, Amsterdam, 1066 CX, Neth.
SOURCE: Nature (London) (2000), 404(6775), 296-298
CODEN: NATUAS; ISSN: 0028-0836
PUBLISHER: Nature Publishing Group
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Originally discovered in plants, the phenomenon of co-suppression by transgenic DNA has since been obsd. in many organisms from fungi to animals: introduction of transgenic copies of a gene results in reduced expression of the transgene as well as the endogenous gene. The effect depends on sequence identity between transgene and endogenous gene. Some cases of co-suppression resemble RNA interference (the exptl. silencing of genes by the introduction of double-stranded RNA), as RNA seems to be both an important initiator and a target in these processes. Here we show that co-suppression in Caenorhabditis **elegans** is also probably mediated by RNA mols. Both RNA interference and co-suppression have been implicated in the silencing of transposons. We now report that mutants of C. **elegans** that are defective in transposon silencing and RNA interference (mut-2, mut-7, mut-8 and mut-9) are in addn. resistant to co-suppression. This indicates that RNA interference and co-suppression in C. **elegans** may be mediated at least in part by the same mol. machinery, possibly through RNA-guided degrdn. of mRNA mols.
REFERENCE COUNT: 30
REFERENCE(S): (2) Baulcombe, D; Curr Opin Biotechnol 1996, V7, P173 CAPLUS
(3) Cogoni, C; Nature 1999, V399, P166 CAPLUS
(4) Collins, J; Nature 1987, V328, P726 CAPLUS
(6) Fire, A; Trends Genet 1999, V15, P358 CAPLUS
(7) Francis, R; Genetics 1995, V139, P579 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L8 ANSWER 6 OF 10 LIFESCI COPYRIGHT 2001 CSA
ACCESSION NUMBER: 2000:70572 LIFESCI
TITLE: Double-Stranded RNA as a Template for Gene Silencing
AUTHOR: Bass, B.L.
CORPORATE SOURCE: Department of Biochemistry and Howard Hughes Medical Institute, University of Utah School of Medicine, Salt Lake City, UR 84132, USA; E-mail: bbass@howard.genetics.utah.edu
SOURCE: Cell, (20000428) vol. 101, no. 3, pp. 235-238. ISSN: 0092-8674.
DOCUMENT TYPE: Journal
TREATMENT CODE: General Review
FILE SEGMENT: G; N
LANGUAGE: English

AB When double-stranded RNA (dsRNA) corresponding to a sense and antisense sequence of an endogenous mRNA is introduced into a cell, in organisms ranging from trypanosomes to mice, the cognate mRNA is degraded and the gene is silenced. This type of posttranscriptional gene silencing (PTGS) was first discovered in C. **elegans** and is called RNA interference, or RNAi. RNAi shows many similarities to the PTGS that is sometimes observed when a transgene is introduced into a cell, and the processes seem to require some of the same gene products. If transgene-induced silencing of an endogenous gene, or **cosuppression**, also involves dsRNA, somehow the cell must make both sense and antisense copies of the transgene sequence. PTGS has captured the interest (and imagination) of geneticists and molecular biologists alike, and now the first clues about its mechanism will certainly bring the biochemists into the fold. As is often the case for biological processes, the first hint about the mechanism comes from the identification of molecules that appear to be reaction intermediates. In particular, several recent papers report the identification of small RNA molecules, 21-25 nucleotides in length (21- to 25-mers), that correspond to sense and antisense pieces of the dsRNA or transgene introduced into the cell.

L8 ANSWER 7 OF 10 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.
ACCESSION NUMBER: 1999054363 EMBASE
TITLE: Less from more: **Cosuppression** of transposable elements.
AUTHOR: Birchler J.A.; Pal-Bhadra M.; Bhadra U.
CORPORATE SOURCE: J.A. Birchler, Division of Biological Sciences, University of Missouri, Columbia, MO 65211-7400, United States. birchler@biosci.mbp.missouri.edu
SOURCE: Nature Genetics, (1999) 21/2 (148-149). Refs: 16 ISSN: 1061-4036 CODEN: NGENEC
COUNTRY: United States
DOCUMENT TYPE: Journal; (Short Survey)
FILE SEGMENT: 022 Human Genetics
LANGUAGE: English

L8 ANSWER 8 OF 10 LIFESCI COPYRIGHT 2001 CSA DUPLICATE 3
ACCESSION NUMBER: 1999:50320 LIFESCI
TITLE: RNAi and double-strand RNA
AUTHOR: Sharp, P.A.
CORPORATE SOURCE: Center for Cancer Research and Department of Biology, Massachusetts Institute of Technology, Cambridge, MA 02139-4307, USA; E-mail: sharppa@mit.edu
SOURCE: Genes & Development [Genes Dev.], (19990115) vol. 13, no. 2, pp. 139-141. ISSN: 0890-9369.
DOCUMENT TYPE: Journal
TREATMENT CODE: General Review
FILE SEGMENT: N
LANGUAGE: English

AB Double-strand RNA (dsRNA) is a signal for gene-specific silencing of expression in a number of organisms. This phenomenon was demonstrated recently in Caenorhabditis **elegans** when dsRNA was injected into the worm and the corresponding gene products disappeared from both the somatic cells of the organism as well as in its F sub(1) progeny. This RNA interference, RNAi, has been generalized to many genes in C. **elegans**. ds-RNA can also suppress expression of specific genes in plants, a component of the phenomenon called **cosuppression**. Two recent reports document dsRNA-mediated interference with expression of specific genes in other organisms. Double-strand RNA produced gene-specific phenotypes in Trypanosoma brucei and, very recently, dsRNA-mediated interference was demonstrated in Drosophila. Thus, the RNAi phenomenon is likely to be a general mechanism for gene regulation and may be critical for many developmental and antiviral processes.

L8 ANSWER 9 OF 10 MEDLINE DUPLICATE 4
ACCESSION NUMBER: 1999061928 MEDLINE
DOCUMENT NUMBER: 99061928
TITLE: Double-stranded RNA induces mRNA degradation in Trypanosoma brucei.
AUTHOR: Ngo H; Tschudi C; Gull K; Ullu E
CORPORATE SOURCE: Department of Internal Medicine, Yale University School of Medicine, 333 Cedar Street, New Haven, CT 06520-8022, USA.
CONTRACT NUMBER: AI28798 (NIAID)
SOURCE: PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA, (1998 Dec 8) 95 (25) 14687-92. Journal code: PV3. ISSN: 0027-8424.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Cancer Journals
ENTRY MONTH: 199903
ENTRY WEEK: 19990303
AB Double-stranded RNA (dsRNA) recently has been shown to give rise to genetic interference in Caenorhabditis **elegans** and also is likely to be the basis for phenotypic **cosuppression** in plants in certain instances. While constructing a plasmid vector for transfection of trypanosome cells, we serendipitously discovered that in vivo expression of dsRNA of the alpha-tubulin mRNA 5' untranslated region (5' UTR) led to multinucleated cells with striking morphological alterations and a specific block of cytokinesis. Transfection of synthetic alpha-tubulin 5' UTR dsRNA, but not of either strand individually, caused the same phenotype. On dsRNA transfection, tubulin mRNA, but not the corresponding pre-mRNA, was rapidly and specifically degraded, leading to a deficit of alpha-tubulin synthesis. The transfected cells were no longer capable of carrying out cytokinesis and eventually died. Analysis of cytoskeletal structures from these trypanosomes revealed defects in the microtubules of the flagellar axoneme and of the flagellar attachment zone, a complex cortical structure that we propose is essential for establishing the path of the cleavage furrow at cytokinesis. Last, dsRNA-mediated mRNA degradation is not restricted to alpha-tubulin mRNA but can be applied to other cellular mRNAs, thus establishing a powerful tool to genetically manipulate these important protozoan parasites.

L8 ANSWER 10 OF 10 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1998:465069 CAPLUS
DOCUMENT NUMBER: 129:184695
TITLE: Double-stranded RNA as a mediator in sequence-specific

genetic silencing and co-suppression
AUTHOR(S): Montgomery, Mary K.; Fire, Andrew
CORPORATE SOURCE: Dep. Embryology, Carnegie Inst. Washington, Baltimore,
MD, 21210, USA
SOURCE: Trends Genet. (1998), 14(7), 255-258
CODEN: TRGEE2; ISSN: 0168-9525
PUBLISHER: Elsevier Science Ltd.
DOCUMENT TYPE: Journal; General Review
LANGUAGE: English
AB A review and discussion with 24 refs. on the possibility that double-stranded RNA (dsRNA), rather than sense or antisense single-stranded RNAs alone, is the effector mol. responsible for RNA-mediated silencing and co-suppression. Topics include: RNA-mediated genetic interference in nematode; RNA-mediated silencing and co-suppression in plants; possible mechanisms for RNA-mediated interference; and RNA-mediated interference mechanisms in organisms other than nematodes and plants.

=> s rnai

L9 639 RNAI

=> s l9 and human

L10 85 L9 AND HUMAN

=> dup rem l10

PROCESSING COMPLETED FOR L10
L11 57 DUP REM L10 (28 DUPLICATES REMOVED)

=> d l11 ibib abs 50-57

L11 ANSWER 50 OF 57 MEDLINE
ACCESSION NUMBER: 81267452 MEDLINE
DOCUMENT NUMBER: 81267452
TITLE: Inter-RNA homology and possible roles of small RNAs.
AUTHOR: Gojobori T; Nei M
SOURCE: JOURNAL OF MOLECULAR EVOLUTION, (1981) 17 (4) 245-50.
Journal code: J76. ISSN: 0022-2844.
PUB. COUNTRY: GERMANY, WEST: Germany, Federal Republic of
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198112
AB The nucleotide sequence of a segment of U1 and U3b small RNAs (sRNAs) is shown to have a high complementarity with the nucleotide sequence of a part of the leader region of almost all eukaryotic genes studied so far. The complementary region of U3b is located in the unpaired segment of the secondary structure of U3b constructed by Reddy et al. (1979). A similar complementarity is also observed between these RNAs and the leader regions of eukaryotic viruses, but the complementary region is not always identical with that for eukaryotic genes. Complementarity is also observed between the 3' end of 18S rRNA and a segment of U1 or U3b which is almost contiguous to the region complementary with mRNA. These observations suggest that U1 and U3b may be involved in mRNA processing and transport in the nucleus or in translation in the cytoplasm. In addition to U1 and U3b, another sRNA, i.e., 4.5S **RNAI**, is shown to have segments which are homologous to the Hogness box of the flanking region of gene and the Proudfoot-Brownlee (PB) box of mRNA near the poly(A) attachment site.

The two segments which are complementary with these boxes are located almost contiguously on a co-joined loop of the secondary structure of 4.5S
**RNAI** constructed by Ro-Choi et al. (1972). Since the Hogness box and PB box are both considered as a recognition site by the RNA polymerase, it is possible that 4.5S **RNAI** is involved in mediating gene transcription.

L11 ANSWER 51 OF 57 MEDLINE
ACCESSION NUMBER: 81244772 MEDLINE
DOCUMENT NUMBER: 81244772
TITLE: Nucleotide sequence complementarity between adenovirus 2-coded VA RNA and host cell pre-mRNA. A possible regulatory mechanism of cellular RNA splicing by VA RNA.
AUTHOR: Naora H; Deacon N J
SOURCE: MOLECULAR BIOLOGY REPORTS, (1981 May 22) 7 (1-3) 115-21. Journal code: NGW. ISSN: 0301-4851.
PUB. COUNTRY: Netherlands
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198111
AB Using a computer program, complementary of nucleotide sequences was assessed between adenovirus 2-coded VA RNA and presumptive cellular and viral 'pre-mRNAs'. In this paper, the possibility is considered that the splicing of cellular 'pre-mRNA' can be regulated in such a way that the formation of the proper intramolecular double-stranded hairpin structures,
key elements for RNA splicing, is prevented by the binding of VA **RNAI** or RNAII to the nucleotide sequences around the exon-intron and intron-exon joint sites of cellular 'pre-mRNA' molecules. Complementarity assessment showed that VA **RNAI** can bind to the joint sites in such a way as to form an omega shape at two separate regions around the joint sites of cellular 'pre-mRNA'. Whereas VA **RNAI** is not capable of binding to viral hexon 'pre-mRNA' in the same manner as it does to cellular 'pre-mRNA', the binding may occur in a different way. Such differential binding is discussed in relation to the post-transcriptional sequence selection which takes place during the late phase of adenovirus infection.

L11 ANSWER 52 OF 57 MEDLINE DUPLICATE 16
ACCESSION NUMBER: 81069892 MEDLINE
DOCUMENT NUMBER: 81069892
TITLE: Multiple factors are required for the accurate transcription of purified genes by RNA polymerase III.
AUTHOR: Segall J; Matsui T; Roeder R G
CONTRACT NUMBER: CA16640 (NCI)
CA23615 (NCI)
P30 CA 176217 (NCI)
SOURCE: JOURNAL OF BIOLOGICAL CHEMISTRY, (1980 Dec 25) 255 (24) 11986-91. Journal code: HIV. ISSN: 0021-9258.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198104
AB Cell-free extracts (S-100) prepared from cultured mammalian KB cells have previously been shown to direct accurate and selective transcription of class III genes by RNA polymerase III. We have fractionated the KB S-100 and have found that multiple components are essential for the accurate transcription of these genes. After the S-100 has been separated into four
different protein fractions by chromatography on phosphocellulose, two fractions are required, in addition to RNA polymerase III, for active and selective transcription of the virus-associated **RNAI** gene of

adenovirus 2 and a tRNA gene; a third fraction is required, along with these components, for the reconstitution of 5 S RNA gene transcription. At least two of these components are distinct from the four factors required for accurate initiation of transcription by RNA polymerase II (Matsui, T., Segall, J., Weil, P. A., and Roeder, R. G. (1980) J. Biol. Chem. 255, 11992-11996).

L11 ANSWER 53 OF 57 MEDLINE
ACCESSION NUMBER: 80234635 MEDLINE
DOCUMENT NUMBER: 80234635
TITLE: Structure of genes for virus-associated **RNAI** and RNAII of adenovirus type 2.
AUTHOR: Akusjarvi G; Mathews M B; Andersson P; Vennstrom B; Pettersson U
SOURCE: PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA, (1980 May) 77 (5) 2424-8. Journal code: PV3. ISSN: 0027-8424.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-J01917; GENBANK-J01918; GENBANK-J01919; GENBANK-J01920; GENBANK-J01921; GENBANK-J01922; GENBANK-J01923; GENBANK-J01924; GENBANK-J01925; GENBANK-J01926; GENBANK-J01927; GENBANK-J01928; GENBANK-J01929; GENBANK-J01930; GENBANK-J01931; GENBANK-J01932; GENBANK-J01933; GENBANK-J01934; GENBANK-J01935; GENBANK-J01936; GENBANK-J01937; GENBANK-J01938; GENBANK-J01939; GENBANK-J01940; GENBANK-J01941; GENBANK-J01942; GENBANK-J01943; GENBANK-J01944; GENBANK-J01945; GENBANK-J01946; +
ENTRY MONTH: 198011
AB A DNA sequence, 552 base pairs in length, encoding the two "virus-associated" (VA) RNAs of adenovirus type 2 is presented. Comparison of the oligonucleotide maps of VA **RNAI** and VA RNII with the established sequence permits identification of the genes for these RNAs. VA **RNAI** is 157-160 nucleotides long and VA RNAII 158-163 nucleotides long, depending on the exact length of their heterogeneous 3' end. The genes are separated by a spacer of about 98 nucleotides. The RNAs exhibit scattered regions of primary sequence homology and can adopt secondary structures which resemble each other closely in their configuration and stability. VA RNAII is also capable of assuming a different configuration that is energetically more favorable. The data suggest that the two RNA genes may have arisen by duplication of an ancestral gene and that the folding of the RNA chain may be of importance for the function of VA RNAs. Hypothetical RNA polymerase III recognition sequences and the coding potential of the region are discussed.

L11 ANSWER 54 OF 57 MEDLINE DUPLICATE 17
ACCESSION NUMBER: 79194183 MEDLINE
DOCUMENT NUMBER: 79194183
TITLE: Faithful transcription of eukaryotic genes by RNA polymerase III in systems reconstituted with purified DNA templates.
AUTHOR: Weil P A; Segall J; Harris B; Ng S Y; Roeder R G
SOURCE: JOURNAL OF BIOLOGICAL CHEMISTRY, (1979 Jul 10) 254 (13) 6163-73. Journal code: HIV. ISSN: 0021-9258.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals

ENTRY MONTH: 197910

AB The virus-associated (VA) **RNAI** gene in **human** adenovirus 2 DNA has been shown by Wu (Wu, G. J. (1978) Proc. Natl. Acad. Sci. U. S. A. 75, 2175--2179) to be transcribed by RNA polymerase III in a **human** KB cell-free extract. In the present report we have examined the fidelity of transcription of adenovirus 2 DNA and Xenopus oocyte 5 S DNA templates by RNA polymerase III in extracts derived from cultured **human**, murine, and amphibian kidney cells, Size and sequences analysis of the discrete transcripts synthesized in these homologous and heterologous systems indicate that they result from accurate transcription of the corresponding genes. The specific transcripts identified include both the adenovirus VA **RNAI** and VA RNAII, Xenopus 5 S RNA, and VA **RNAI** and 5 S RNA species with elongated 3' termini. The extracts derived from the various cell types differ in the ability to discriminate between the two VA RNA genes or between the heterogeneous 5 S RNA genes in the cloned DNA fragment. Wherease the **human** cell extracts transcribe the VA **RNAI** and VA RNAII genes of adenovirus at a relative frequency close to that observed in isolated nuclei, the amphibian cell extract appears to transcribe only the VA **RNAI** gene. The amphibian cell extract transcribes primarily that 5 S RNA gene (within 5 S DNA) which encodes the dominant oocyte 5 S RNA, whereas the **human** cell extract transcribes at least two distinct 5 S RNA genes. Additionally, it is shown that the VA **RNAI** and VA RNAII genes have separate promotor sites. The kinetics of the transcription reactions have been examined and conditions optimal for specific transcription have been established by examining the effects of salt, metal ion, and template concentrations on both total and specific RNA synthesis. It is also shown that components in the cell-free extract (from **human** cells) are active in directing the accurate transcription of adenovirus DNA by purified RNA polymerase III.

L11 ANSWER 55 OF 57 BIOSIS COPYRIGHT 2001 BIOSIS

ACCESSION NUMBER: 1978:252140 BIOSIS

DOCUMENT NUMBER: BA66:64637

TITLE: STRUCTURAL RELATIONSHIPS OF LOW MOLECULAR WEIGHT VIRAL RNA SYNTHESIZED BY RNA POLYMERASE III IN NUCLEI FROM ADENOVIRUS 2 INFECTED CELLS.

AUTHOR(S): HARRIS B; ROEDER R G

CORPORATE SOURCE: DIV. BIOL. BIOMED. SCI., DEP. BIOL. CHEM., WASHINGTON UNIV., ST. LOUIS, MO. 63110, USA.

SOURCE: J BIOL CHEM, (1978) 253 (12), 4120-4127.
CODEN: JBCHA3. ISSN: 0021-9258.

FILE SEGMENT: BA; OLD

LANGUAGE: English

AB Previous studies showed that endogenous class III RNA polymerase(s) in nuclei from adenovirus 2-infected [**human** oral carcinoma KB] cells synthesize virus-coded RNA species which are approximately 200 (V200), 156 (V156) and 140 (V140) nucleotides in length. The V156 nuclear RNA is identical in sequence to the major virus-associated RNA (VA RNA1 or 5.5 S RNA) synthesized in intact cells. The V140 RNA contains several components, one of which appears identical to a minor virus-associated RNA (VA RNAII) which is synthesized in infected cells. Thus transcription of the VA RNA1 and VA RNAII genes in vitro accurately reflects the in vivo transcription of these genes. The V200 RNA contains all the nucleotide sequences found in V156 RNA plus an additional 38-40 nucleotides on the 3' terminus. Transcription of the gene encoding this RNA species terminates within a stretch of 6 deoxythymidylic acid residues which are located 38 nucleotides beyong the predicted termination site for VA **RNAI**

and which are preceded by a GC-rich sequence of nucleotides. Either the V200 RNA is a precursor to the VA **RNAI** or the RNA polymerase III occasionally reads through the presumptive VA **RNAI** gene termination signal and stops at a potentially stronger downstream termination site.

L11 ANSWER 56 OF 57 BIOSIS COPYRIGHT 2001 BIOSIS
ACCESSION NUMBER: 1978:206086 BIOSIS
DOCUMENT NUMBER: BA66:18583
TITLE: THE LOW MOLECULAR WEIGHT OF RNA OF ADENOVIRUS 2 INFECTED CELLS.
AUTHOR(S): MATHEWS M B; PETTERSSON U
CORPORATE SOURCE: COLD SPRING HARBOR LAB., P.O. BOX 100, COLD SPRING HARBOR, N.Y., USA.
SOURCE: J MOL BIOL, (1978) 119 (2), 293-328.
CODEN: JMOBAK. ISSN: 0022-2836.
FILE SEGMENT: BA; OLD
LANGUAGE: English
AB The cytoplasm of [**human** cervical carcinoma] HeLa cells infected with adenovirus type 2 contains many species of low MW RNA, including several of viral origin. Besides a 9 S mRNA, the viral genome gives rise to 2 spp. of virus-associated RNA: the major species is 5.5 S RNA or virus-associated **RNAI**, and the minor species is 5.2 S RNA or virus-associated RNAII. Virus-associated **RNAI** occurs in the cytoplasm in several electrophoretically separable forms, and its sequences are also present in high MW nuclear RNA but not in cytoplasmic mRNA. The structure of virus-associated RNAII distinct from that of the major species, and the position of its gene is mapped on the viral genome.
The 2 virus-associated RNA genes are located on the r strand near position
30 of the adenovirus type 2 physical map, and are separated by a spacer of
about 75 base-pairs.

L11 ANSWER 57 OF 57 BIOSIS COPYRIGHT 2001 BIOSIS
ACCESSION NUMBER: 1977:132611 BIOSIS
DOCUMENT NUMBER: BA63:27475
TITLE: CHARACTERISTICS OF SKELETAL MUSCULAR TISSUE RIBOSOMES.
AUTHOR(S): KHOROSHKOV YU A; SHISHKIN S S
SOURCE: BYULL EKSP BIOL MED, (1976) 81 (5), 534-536.
CODEN: BEBMAE. ISSN: 0365-9615.
FILE SEGMENT: BA; OLD
LANGUAGE: Unavailable
AB EM and chemical analysis of the fraction of RNP [ribonucleoprotein] particles from samples of the muscle tissue of the posterior limb of rat and of the **human** musculus rectus abdominis were carried out to characterize ribosomes of the skeletal-muscular tissue. The RNP particle fraction contained functionally active mono- and polyribosomes. Two RNA fractions, **RNAI** and RNAIII, were isolated. **RNAI** represents a set of ribosomic RNA with sedimentation coefficients 26-28S [Svedberg unit], 16-18S and 4-5S. RNAIII contained no ribosomal RNA, and by its nucleotide composition was affiliated to the nucleotide composition
of rat DNA. The structural organization of ribosomes in the cytoplasm of the muscular fibers corresponded to the pictures observed in the RNP particle fraction. Polyribosomes are determined in the regions of physiological regeneration of myofibrillae and represent complexes consisting of 5 and more monoribosomes arranged like beads. Ribosome-like particles escape through the nuclear membrane; this fact and the chemical analysis data indicated the periodic passage of RNA from the nucleus into the cytoplasm of the muscle fiber.

=> d history

(FILE 'HOME' ENTERED AT 18:00:34 ON 08 JAN 2001)

FILE 'MEDLINE, CAPLUS, LIFESCI, EMBASE, BIOSIS' ENTERED AT 18:00:58 ON 08
JAN 2001
L1 26433 S RNA AND ANTISENSE
L2 26594 S DSRNA OR (DOUBLE(S) STRANDED(S) RNA)
L3 698 S L2 AND ANTISENSE
L4 37 S L3 AND TRIPLEX
L5 18 DUP REM L4 (19 DUPLICATES REMOVED)
L6 291 S COSUPPRESSION
L7 20 S L6 AND ELEGANS
L8 10 DUP REM L7 (10 DUPLICATES REMOVED)
L9 639 S RNAI
L10 85 S L9 AND HUMAN
L11 57 DUP REM L10 (28 DUPLICATES REMOVED)

=> s l6 and human

L12 13 L6 AND HUMAN

=> dup rem l12

PROCESSING COMPLETED FOR L12
L13 6 DUP REM L12 (7 DUPLICATES REMOVED)

=> d l13 ibib abs tot

L13 ANSWER 1 OF 6 MEDLINE
ACCESSION NUMBER: 1999140758 MEDLINE
DOCUMENT NUMBER: 99140758
TITLE: Less from more: **cosuppression** of transposable elements [news; comment].
COMMENT: Comment on: Nat Genet 1999 Feb;21(2):209-12
AUTHOR: Birchler J A; Pal-Bhadra M; Bhadra U
SOURCE: NATURE GENETICS, (1999 Feb) 21 (2) 148-9.
Journal code: BRO. ISSN: 1061-4036.
PUB. COUNTRY: United States
Commentary
News Announcement
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199904

L13 ANSWER 2 OF 6 MEDLINE DUPLICATE 1
ACCESSION NUMBER: 1999251536 MEDLINE
DOCUMENT NUMBER: 99251536
TITLE: RESPONSIVE-TO-ANTAGONIST1, a Menkes/Wilson disease-related copper transporter, is required for ethylene signaling in Arabidopsis.
AUTHOR: Hirayama T; Kieber J J; Hirayama N; Kogan M; Guzman P; Nourizadeh S; Alonso J M; Dailey W P; Dancis A; Ecker J R
CORPORATE SOURCE: Plant Science Institute, Department of Biology, University of Pennsylvania, Philadelphia 19104, USA.
SOURCE: CELL, (1999 Apr 30) 97 (3) 383-93.
Journal code: CQ4. ISSN: 0092-8674.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Cancer Journals
OTHER SOURCE: GENBANK-AF091112; GENBANK-AF082565
ENTRY MONTH: 199907
ENTRY WEEK: 19990704
AB Ethylene is an important regulator of plant growth. We identified an

Arabidopsis mutant, responsive-to-antagonist1 (ran1), that shows ethylene phenotypes in response to treatment with trans-cyclooctene, a potent receptor antagonist. Genetic epistasis studies revealed an early requirement for RAN1 in the ethylene pathway. RAN1 was cloned and found to encode a protein with similarity to copper-transporting P-type ATPases, including the **human** Menkes/Wilson proteins and yeast Ccc2p. Expression of RAN1 complemented the defects of a ccc2delta mutant, demonstrating its function as a copper transporter. Transgenic CaMV 35S::RAN1 plants showed constitutive expression of ethylene responses, due to **cosuppression** of RAN1. These results provide an in planta demonstration that ethylene signaling requires copper and reveal that RAN1 acts by delivering copper to create functional hormone receptors.

L13 ANSWER 3 OF 6 LIFESCI COPYRIGHT 2001 CSA
ACCESSION NUMBER: 1999:82021 LIFESCI
TITLE: RESPONSIVE-TO-ANTAGONIST1, a Menkes/Wilson Disease-Related Copper Transporter, Is Required for Ethylene Signaling in Arabidopsis
AUTHOR: Hirayama, T.; Kieber, J.J.; Hirayama, N.; Kogan, M.; Guzman, P.; Nourizadeh, S.; Alonso, J.M.; Dailey, W.P.; Dancis, A.; Ecker, J.R.
CORPORATE SOURCE: Plant Science Institute, Department of Biology, Department of Chemistry, Department of Medicine, Division of Hematology-Oncology, University of Pennsylvania, Philadelphia, Pennsylvania 19104; E-mail: jecker@atgenome.bio.upenn.edu
SOURCE: Cell, (19990430) vol. 97, no. 3, pp. 363-393. ISSN: 0092-8674.
DOCUMENT TYPE: Journal
FILE SEGMENT: G
LANGUAGE: English
SUMMARY LANGUAGE: English
AB Ethylene is an important regulator of plant growth. We identified an Arabidopsis mutant, responsive-to-antagonist1 (ran1), that shows ethylene phenotypes in response to treatment with trans-cyclooctene, a potent receptor antagonist. Genetic epistasis studies revealed an early requirement for RAN1 in the ethylene pathway. RAN1 was cloned and found to encode a protein with similarity to copper-transporting P-type ATPases, including the **human** Menkes/Wilson proteins and yeast Ccc2p. Expression of RAN1 complemented the defects of a ccc2 Delta mutant, demonstrating its function as a copper transporter. Transgenic CaMV 35S::RAN1 plants showed constitutive expression of ethylene responses, due to **cosuppression** of RAN1. These results provide an in planta demonstration that ethylene signaling requires copper and reveal that RAN1 acts by delivering copper to create functional hormone receptors.

L13 ANSWER 4 OF 6 MEDLINE DUPLICATE 2
ACCESSION NUMBER: 1999362102 MEDLINE
DOCUMENT NUMBER: 99362102
TITLE: Specific inhibition of hepatitis B virus replication by sense RNA.
AUTHOR: zu Putlitz J; Wands J R
CORPORATE SOURCE: Molecular Hepatology Laboratory, Massachusetts General Hospital Cancer Center and Harvard Medical School, Boston 02129, USA.
CONTRACT NUMBER: CA-35711 (NCI)
AA-02169 (NIAAA)
SOURCE: ANTISENSE AND NUCLEIC ACID DRUG DEVELOPMENT, (1999 Jun) 9 (3) 241-52.
Journal code: CJY. ISSN: 1087-2906.

PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199912
ENTRY WEEK: 19991201
AB We describe effects of sense RNA molecules on hepatitis B virus (HBV) replication and antigen synthesis in transiently transfected cells. When certain subgenomic fragments of HBV were expressed as sense RNA together with a replication-competent genome of HBV, they inhibited HBV replication by up to 75% and HBsAg secretion by up to 60%. The corresponding antisense sequences had a 50% inhibitory effect in one case and no effect in another case. The sense RNA species did not inhibit duck hepatitis B virus (DHBV) replication, suggesting specific inhibitory effects. HBV transcript levels were unaltered in the presence of sense RNA species, consistent with an inhibitory effect mediated at the posttranscriptional level. The inhibition of HBV replication by overexpression of sense RNA derived from the viral genome represents an example of sense **cosuppression** of an animal virus.

L13 ANSWER 5 OF 6 MEDLINE DUPLICATE 3
ACCESSION NUMBER: 1998403434 MEDLINE
DOCUMENT NUMBER: 98403434
TITLE: High-resolution mapping of crossovers in **human** sperm defines a minisatellite-associated recombination hotspot.
AUTHOR: Jeffreys A J; Murray J; Neumann R
CORPORATE SOURCE: Department of Genetics, University of Leicester, United Kingdom.. ajj@le.ac.uk
SOURCE: MOLECULAR CELL, (1998 Aug) 2 (2) 267-73.
Journal code: C5E. ISSN: 1097-2765.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199811
ENTRY WEEK: 19981104
AB Little is known about the fine-scale distribution of meiotic crossovers in **human** chromosomes. Methods have therefore been developed for detecting and mapping recombination products directly in **human** sperm DNA. Analysis of crossovers adjacent to the GC-rich minisatellite MS32, which is known to mutate by conversion and crossover within the repeat array, revealed an intense and highly localized recombination hotspot centered upstream of the locus and extending into the beginning of the minisatellite. Allele-specific **cosuppression** of crossovers and repeat instability suggests that the hotspot is responsible for driving repeat turnover at MS32 and thus that minisatellites might evolve as by-products of localized meiotic recombination in the **human** genome.

L13 ANSWER 6 OF 6 LIFESCI COPYRIGHT 2001 CSA
ACCESSION NUMBER: 1998:113359 LIFESCI
TITLE: High-Resolution Mapping of Crossovers in **Human** Sperm Defines a Minisatellite-Associated Recombination Hotspot
AUTHOR: Jeffreys, A.J.; Murray, J.; Neumann, R.
CORPORATE SOURCE: Department of Genetics, University of Leicester, Leicester LE1 7RH, United Kingdom
SOURCE: Mol. Cell, (19980800) vol. 2, no. 2.
ISSN: 1097-4164.

DOCUMENT TYPE: Journal
FILE SEGMENT: G
LANGUAGE: English
SUMMARY LANGUAGE: English
AB Little is known about the fine-scale distribution of meiotic crossovers in **human** chromosomes. Methods have therefore been developed for detecting and mapping recombination products directly in **human** sperm DNA. Analysis of crossovers adjacent to the GC-rich minisatellite MS32, which is known to mutate by conversion and crossover within the repeat array, revealed an intense and highly localized recombination hotspot centered upstream of the locus and extending into the beginning of the minisatellite. Allele-specific **cosuppression** of crossovers and repeat instability suggests that the hotspot is responsible for driving repeat turnover at MS32 and thus that minisatellites might evolve as by-products of localized meiotic recombination in the **human** genome.

=> d history

(FILE 'HOME' ENTERED AT 18:00:34 ON 08 JAN 2001)

FILE 'MEDLINE, CAPLUS, LIFESCI, EMBASE, BIOSIS' ENTERED AT 18:00:58 ON 08 JAN 2001

L1 26433 S RNA AND ANTISENSE
L2 26594 S DSRNA OR (DOUBLE(S) STRANDED(S) RNA)
L3 698 S L2 AND ANTISENSE
L4 37 S L3 AND TRIPLEX
L5 18 DUP REM L4 (19 DUPLICATES REMOVED)
L6 291 S COSUPPRESSION
L7 20 S L6 AND ELEGANS
L8 10 DUP REM L7 (10 DUPLICATES REMOVED)
L9 639 S RNAI
L10 85 S L9 AND HUMAN
L11 57 DUP REM L10 (28 DUPLICATES REMOVED)
L12 13 S L6 AND HUMAN
L13 6 DUP REM L12 (7 DUPLICATES REMOVED)

=> s l3 and inhibition

L14 131 L3 AND INHIBITION

=> dup rem l14

PROCESSING COMPLETED FOR L14
L15 69 DUP REM L14 (62 DUPLICATES REMOVED)

=> d l15 ibib abs 60-69

L15 ANSWER 60 OF 69 MEDLINE DUPLICATE 21
ACCESSION NUMBER: 92126178 MEDLINE
DOCUMENT NUMBER: 92126178
TITLE: The anti-gene strategy: control of gene expression by triplex-forming-oligonucleotides.
AUTHOR: Hel`ene C
CORPORATE SOURCE: Laboratoire de Biophysique, Museum National d'Histoire Naturelle, INSERM U201-CNRS UA 481, Paris, France..
SOURCE: ANTI-CANCER DRUG DESIGN, (1991 Dec) 6 (6) 569-84. Ref: 60
Journal code: AC5. ISSN: 0266-9536.
PUB. COUNTRY: ENGLAND: United Kingdom
Journal; Article; (JOURNAL ARTICLE)

General Review; (REVIEW)
(REVIEW, TUTORIAL)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Cancer Journals
ENTRY MONTH: 199205

AB Oligonucleotides are being developed to selectively inhibit gene expression at the translational level (**antisense** oligonucleotides) and at the transcriptional level (anti-gene oligonucleotides). This review deals with the anti-gene strategy whereby an oligonucleotide binds to the major groove of **double** helical DNA where it forms a local triple helix. The molecular mechanisms for DNA recognition by triple helix formation are discussed together with some of the rules presently available to design the sequence and orientation of the triple helix forming oligonucleotide. Triplex stability can be enhanced by covalent attachment of an intercalating agent to the terminal nucleotide of the oligonucleotide. The intercalating agent can be used to induce irreversible reactions in the target sequence: **double** strand cleavage by a phenanthroline-Cu chelate in the presence of a reducing agent, photo-induced cleavage by ellipticine derivatives, photo-induced cross-linking of the two DNA strands by psoralen... Triple helix-forming oligonucleotides can be used to control gene expression at the transcriptional level. **Inhibition** of binding of transcription activating factors by triplex formation modulates the level of transcription of the target gene. Binding of a triplex-forming oligonucleotide immediately downstream of the **RNA** polymerase binding site can inhibit transcription initiation as shown with the E. coli beta-lactamase gene. Studies with cells in culture show that triple helix formation may occur in the intracellular environment and consequently leads to transcription **inhibition**. This inhibitory effect can be made irreversible by using, e.g., psoralen-substituted oligonucleotides. Oligonucleotides synthesized with the alpha-anomers of nucleotide units are resistant to nucleases and still form triple helices with **double-stranded** DNA. Oligo-[alpha]-deoxynucleotides can be derived by stabilizing (intercalating) agents or reactive groups (cleaving reagents, cross-linkers ...). The results presently available provide a rational basis for the development of new tools for cellular biology and of new therapeutical approaches to selectively control gene expression at the transcriptional level.

L15 ANSWER 61 OF 69 MEDLINE DUPLICATE 22
ACCESSION NUMBER: 91234282 MEDLINE
DOCUMENT NUMBER: 91234282
TITLE: **Inhibition** of heterologous strains of HIV by **antisense** RNA [see comments].
COMMENT: Comment in: AIDS 1991 Feb;5(2):225-6
AUTHOR: Rhodes A; James W
CORPORATE SOURCE: Sir William Dunn School of Pathology, University of Oxford, Berkshire, UK.
SOURCE: AIDS, (1991 Feb) 5 (2) 145-51.
Journal code: AID. ISSN: 0269-9370.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199108

AB **Antisense RNA** can inhibit the expression of messenger RNAs (mRNAs) to which they are complementary by a variety of mechanisms and might provide the basis for antiviral therapies of high selectivity. In a previous study of six retrovirally expressed **antisense** RNAs targeted to HIV-1IIIB, we found that two significantly reduced HIV-1IIIB replication. Here we test the degree to which this inhibitory effect tolerates the natural variation found in the nucleotide sequence of different strains of HIV-1. We show that the longer of the two inhibitory **antisense** RNAs (600 bases) inhibits replication of HIV strains RF, MN and SF2 to at least as great an extent as it does the homologous

strain. In contrast, the shorter (71 bases) does not inhibit replication of the heterologous strains. An examination of the predicted positions of the mismatches in the duplexes formed between the IIIB **antisense** RNAs and the mRNAs of heterologous strains suggests that one requirement of an inhibitory **antisense RNA** is that it can form a perfect duplex with its target mRNA of at least some 51-64 base-pairs. Although the observations presented here are not definitive proof of this, they are reminiscent of the structural requirements deduced for the **double-stranded RNA**-mediated induction of interferon and the activation of interferon-induced 2', 5'-oligo(A) synthetase and protein kinase. We tested the ability of **antisense RNA** to inhibit HIV replication in Jurkat, CEM, U937 and HeLa-T4 cells. The level of **inhibition** of HIV-1IIIB replication varied according to the cell line in which it was expressed, but in all cases was significant.

L15 ANSWER 62 OF 69 MEDLINE DUPLICATE 23
ACCESSION NUMBER: 90223031 MEDLINE
DOCUMENT NUMBER: 90223031
TITLE: Phosphate-methylated DNA aimed at HIV-1 RNA loops and integrated DNA inhibits viral infectivity [retracted by Moody HM, Quaedflieg PJ, Koole LH, van Genderen MH, Buck HM, Smit L, Jurriaans S, Geelen JL, Goudsmit J. In: Science 1990 Oct 5;250(4977):125-6].
AUTHOR: Buck H M; Koole L H; van Genderen M H; Smit L; Geelen J L; Jurriaans S; Goudsmit J
CORPORATE SOURCE: Department of Organic Chemistry, Eindhoven University of Technology, The Netherlands.
SOURCE: SCIENCE, (1990 Apr 13) 248 (4952) 208-12.
Journal code: UJ7. ISSN: 0036-8075.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
(RETRACTED PUBLICATION)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Cancer Journals
ENTRY MONTH: 199007
AB Phosphate-methylated DNA hybridizes strongly and specifically to natural DNA and **RNA**. Hybridization to single-**stranded** and **double-stranded** DNA leads to site-selective blocking of replication and transcription. Phosphate-methylated DNA was used to interrupt the life cycle of the human immunodeficiency virus type-1 (HIV-1), the causative agent of acquired immunodeficiency syndrome (AIDS). Both **antisense** and sense phosphate-methylated DNA 20-nucleotide oligomers, targeted at the transactivator responsive region and the primer binding site, caused complete **inhibition** of viral infectivity at a low concentration. Hybridization of phosphate-methylated DNA with folded and unfolded **RNA** was studied by ultraviolet and proton nuclear magnetic resonance spectroscopy. The combined results of hybridization studies and biological experiments suggest that the design of effective **antisense** phosphate-methylated DNA should focus on hairpin loop structures in the viral **RNA**. For sense systems, the 5' end of the integrated viral genome is considered to be the important target site.

L15 ANSWER 63 OF 69 BIOSIS COPYRIGHT 2001 BIOSIS
ACCESSION NUMBER: 1989:202917 BIOSIS
DOCUMENT NUMBER: BA87:103821
TITLE: ACTIVATION OF INTERFERON-REGULATED **DOUBLE-STRANDED RNA**-DEPENDENT ENZYMES BY HUMAN IMMUNODEFICIENCY VIRUS 1 LEADER **RNA**.

AUTHOR(S): SENGUPTA D; SILVERMAN R H
CORPORATE SOURCE: DEP. PATHOL., UNIFORMED SERV. UNIV. HEALTH SCI., 4301 JONES
BRIDGE RD., BETHESDA, MD. 20814-4799, USA.
SOURCE: NUCLEIC ACIDS RES, (1989) 17 (3), 969-978.
CODEN: NARHAD. ISSN: 0305-1048.
FILE SEGMENT: BA; OLD
LANGUAGE: English

AB Human immunodeficiency virus-1 (HIV-1) leader **RNA**, which contains **double-stranded** regions due to inverted repeats, was shown to activate the **dsRNA**-dependent enzymes associated with the interferon system. HIV-1 leader **RNA** produced in vitro using SP6 **RNA** polymerase was characterized using probes for **antisense** and sense-strand **RNA**. The **RNA** preparation was free from significant levels of **antisense** **RNA**. HIV-1 leader **RNA** was shown to activate **dsRNA**-dependent protein kinase in a cell-free system from interferon-treated HeLa cells. Affinity resins, consisting of HIV-1 leader **RNA** covalently attached to cellulose, immobilized and activated **dsRNA**-dependent protein kinase and 2-5A-synthetase. HIV-1 leader **RNA**, therefore, may be a contributing factor in the mechanism by which interferon inhibits HIV replication.

L15 ANSWER 64 OF 69 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1990:625201 CAPLUS
DOCUMENT NUMBER: 113:225201
TITLE: Technical basis of the **antisense** approach to therapeutics
AUTHOR(S): Levenson, Corey
CORPORATE SOURCE: Cetus Corp., Emeryville, CA, 94608, USA
SOURCE: Adv. Appl. Biotechnol. Ser. (1989), 2(Discoveries Antisense Nucleic Acids), 15-20
CODEN: AASEE6
DOCUMENT TYPE: Journal; General Review
LANGUAGE: English

AB A review with no refs. Nucleic acids have a capability for self-recognition that is responsible for many of their unique properties. Under appropriate conditions, single-**stranded** DNA and **RNA** mols. bind to their complementary strands (mirror images in terms of nucleotide sequence) to form stable **double-stranded** structures (helical duplexes). The specificity of this interaction allows nucleic acids to be utilized as sensitive diagnostic probes and is the basis of the **antisense** mechanism of genetic regulation. For nucleic acids to be replicated, or for the information contained therein to be translated into essential proteins, it is necessary for the DNA or RNA to exist, at least transiently, in single-stranded form. It is while they are in this single-stranded form that they are susceptible to **inhibition** by hybridization with complementary single-stranded nucleic acids.

L15 ANSWER 65 OF 69 MEDLINE DUPLICATE 24
ACCESSION NUMBER: 88124934 MEDLINE
DOCUMENT NUMBER: 88124934
TITLE: **Antisense RNA** inhibits endogenous gene expression in mouse preimplantation embryos: lack of **double-stranded RNA** "melting" activity.
AUTHOR: Bevilacqua A; Erickson R P; Hieber V
CORPORATE SOURCE: Department of Human Genetics, University of Michigan Medical School, Ann Arbor 48109-0618.
CONTRACT NUMBER: HD 20670 (NICHD)
SOURCE: PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA, (1988 Feb) 85 (3) 831-5.
Journal code: PV3. ISSN: 0027-8424.
PUB. COUNTRY: United States

Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Cancer Journals
ENTRY MONTH: 198805

AB beta-Glucuronidase activity increases 60-fold from the 4-cell to the blastocyst stage during in vitro development of mouse preimplantation embryos, secondary to a 13-fold increase in beta-glucuronidase mRNA. Injections of **antisense** RNA from a beta-glucuronidase cDNA clone lacking the 5'-untranslated region and the coding sequences for approximately equal to 150 N-terminal amino acids were effective in partially blocking the appearance of beta-glucuronidase activity. Injection of the same RNA, capped with guanosine(5')triphospho(5')guanosin e (GpppG), into each blastomere at the 4-cell stage yielded 75% **inhibition** of enzyme activity at the blastocyst stage. Injections of the sense strand or of an unrelated RNA did not alter the normal increase in activity of the enzyme. These results are in accord with our inability to detect RNA-duplex "melting" activity in 1-cell mouse embryos.

We suggest that it may be possible to analyze genetics of mammalian development by **antisense** techniques.

L15 ANSWER 66 OF 69 MEDLINE DUPLICATE 25
ACCESSION NUMBER: 88296476 MEDLINE
DOCUMENT NUMBER: 88296476
TITLE: Excess **antisense** RNA from infectious recombinant SV40 fails to inhibit expression of a transfected, interferon-inducible gene.
AUTHOR: Kerr S M; Stark G R; Kerr I M
CORPORATE SOURCE: Imperial Cancer Research Fund Laboratories, London, England.
SOURCE: EUROPEAN JOURNAL OF BIOCHEMISTRY, (1988 Jul 15) 175 (1) 65-73.
Journal code: EMZ. ISSN: 0014-2956.
PUB. COUNTRY: GERMANY, WEST: Germany, Federal Republic of
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Cancer Journals
ENTRY MONTH: 198811

AB SV40-based infectious virus constructs were used to produce a high copy number of full-length **antisense RNA** in essentially every cell in a population. Chloramphenicol acetyltransferase (CAT) cDNA was placed in either the sense or **antisense** orientation relative to the SV40 early promoter in helper-free recombinant virus. **RNA** synthesized at high levels from the **antisense** virus was without effect on the expression of a stably-transfected CAT mini-gene controlled by an interferon-inducible promoter in monkey CV1 and large T antigen-expressing tsCOS cells. In **double** infection experiments the **antisense RNA** was similarly without effect on expression from CAT cDNA placed in the sense orientation in a second virus vector. No activation of the ppp(A2'p)nA(n greater than or equal to 2) system was observed after interferon treatment in either type of experiment. There was no evidence, therefore, for the formation of **double-stranded** (ds)**RNA**. It can be concluded that a large excess of a full-length **antisense RNA** is not necessarily sufficient to cause **inhibition** of gene expression even when interferon treatment is used to enhance any effect of **dsRNA**.

L15 ANSWER 67 OF 69 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1987:491168 CAPLUS
DOCUMENT NUMBER: 107:91168
TITLE: Control of gene expression using **antisense** RNA

AUTHOR(S): Tokuhisa, Takeshi
CORPORATE SOURCE: Div. Immunol. Res., Chiba Univ., Chiba, Japan
SOURCE: Jikken Igaku (1987), 5(4), 347-9
CODEN: JIIGEF
DOCUMENT TYPE: Journal
LANGUAGE: Japanese
AB The specific regulation of gene expression by **antisense** RNA is briefly explained. Its principle is the formation of a **double-stranded RNA** via complementary binding of an **antisense RNA** to its sense **RNA**. As examples, the specific inhibitions of the expression of the thymidine kinase gene in mouse L cells, the c-fos gene in NIH3T3 cells, and the Ia-antigen gene in lymphoma M12.4 cells by the resp. **antisense** RNAs are described. The **antisense** RNA will serve as a tool for study of the role of biol. substances during developmental or differential processes.

L15 ANSWER 68 OF 69 MEDLINE
ACCESSION NUMBER: 87041509 MEDLINE
DOCUMENT NUMBER: 87041509
TITLE: Stable repression of ribosomal protein L1 synthesis in Xenopus oocytes by microinjection of **antisense** RNA.
AUTHOR: Wormington W M
CONTRACT NUMBER: HD17691 (NICHD)
SOURCE: PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA, (1986 Nov) 83 (22) 8639-43.
Journal code: PV3. ISSN: 0027-8424.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Cancer Journals
ENTRY MONTH: 198702
AB The synthesis of an endogenous ribosomal protein, L1, is selectively and efficiently inhibited by microinjection of **antisense** L1 RNAs into Xenopus oocytes. Repression of L1 synthesis is achieved within 12 hr and is maintained for 48 hr. RNase-protection assays reveal the formation of RNA X RNA duplexes in vivo between the endogenous L1 mRNA and injected **antisense** transcripts. Partial-length **antisense** RNAs, complementary to only the 3'-terminal region of L1 mRNA, repress translation as effectively as a full-length **antisense** RNA, indicating that complementarity to the 5' region of L1 mRNA is not required for efficient **inhibition**. The use of **antisense** RNA to repress synthesis of an endogenous ribosomal protein provides a functional basis for determining mechanisms that integrate ribosomal protein synthesis with ribosome assembly during oogenesis.

L15 ANSWER 69 OF 69 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1985:607748 CAPLUS
DOCUMENT NUMBER: 103:207748
TITLE: Regulation of the expression of HBV genes
AUTHOR(S): Acs, G.; Price, P.; Sells, M. A.; Zelent, A. Z.; Christman, J. K.
CORPORATE SOURCE: Dep. Biochem., Mount Sinai Sch. Med., New York, NY, 10029, USA
SOURCE: Falk Symp. (1985), 39(Hepatology), 119-24
CODEN: FASYDI; ISSN: 0161-5580
DOCUMENT TYPE: Journal
LANGUAGE: English
AB In cells infected by hepatitis B virus (HBV), the expression of HBV genes may be suppressed by 3 mechanisms. HBV-infected mouse 3T3 cells with integrated HBV DNA were examd. Firstly, the HBV DNA was not methylated at HpaII sites in cells which produced HBV surface antigen and e antigen (HBeAg), but the HBV DNA was methylated at those sites in cells which did not produce the 2 antigens. Thus, HBV gene expression could be suppressed

by DNA methylation. Secondly, the HBV core antigen (HBcAg) was converted into HBeAg by proteolytic enzymes. Thus, by conversion of core protein to a deriv. that can no longer form organized particles, cells could prevent the formation of the nucleoprotein of the virus. Thirdly, **double -stranded RNA**, which hybridized with HBV DNA, was detected in the infected cells. Thus, cells could suppress HBV gene translation with RNAs that anneal with HBV mRNAs.

=> d l15 ibib abs 1-20

L15 ANSWER 1 OF 69 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 2000:756847 CAPLUS
DOCUMENT NUMBER: 133:318250
TITLE: **Double-stranded RNA** for the post-transcriptional **inhibition** of gene expression and its therapeutic uses
INVENTOR(S): Pachuk, Catherine; Satishchandran, C.
PATENT ASSIGNEE(S): American Home Products Corp., USA
SOURCE: PCT Int. Appl., 64 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|---|---|---|---|---|
| WO 2000063364 | A2 | 20001026 | WO 2000-US10555 | 20000419 |

W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
RW: GH, GM, KE, LS, MW, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG

PRIORITY APPLN. INFO.: US 1999-130377 19990421
AB A therapeutic compn. for inhibiting the function of a target polynucleotide sequence in a mammalian cell includes an agent that provides to a mammalian cell an at least partially **double-stranded RNA** (**dsRNA**) comprising a polynucleotide sequence of at least about 200 nucleotides in length, said polynucleotide sequence being substantially homologous to a target polynucleotide sequence. This RNA mol. desirably does not produce a functional protein. The agents useful in the compn. can be RNA mols. made by enzymic synthetic methods or chem. synthetic methods in vitro; or made in recombinant cultures of microorganisms and isolated therefrom, or alternatively, can be capable of generating the desired RNA mol. in vivo after delivery to the mammalian cell. In methods of treatment of prophylaxis of virus infections, other pathogenic infections or certain cancers, these compns. are administered in amts. effective to reduce or inhibit the function of the target polynucleotide sequence, which can be of pathogenic origin or produced in response to a tumor or other cancer, among other sources. Use of **dsRNA** to inhibit synthesis of HIV-1 p24 reverse transcriptase is demonstrated. The effect was specific to **dsRNA** derived from the gag gene and the **dsRNA** was more effective than the sense or **antisense** strand alone.

L15 ANSWER 2 OF 69 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.
ACCESSION NUMBER: 2000400553 EMBASE
TITLE: Selective reduction of dormant maternal mRNAs in mouse

oocytes by RNA interference.
AUTHOR: Svoboda P.; Stein P.; Hayashi H.; Schultz R.M.
CORPORATE SOURCE: R.M. Schultz, Department of Biology, University of Pennsylvania, Philadelphia, PA 19104-6018, United States. rschultz@mail.sas.upenn.edu
SOURCE: Development, (2000) 127/19 (4147-4156).
Refs: 59
ISSN: 0950-1991 CODEN: DEVPED
COUNTRY: United Kingdom
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 021 Developmental Biology and Teratology
LANGUAGE: English
SUMMARY LANGUAGE: English
AB Specific mRNA degradation mediated by **double-stranded RNA** (**dsRNA**), which is termed **RNA** interference (RNAi), is a useful tool with which to study gene function in several systems. We report here that in mouse oocytes, RNAi provides a suitable and robust approach to study the function of dormant maternal mRNAs. Mos (originally known as c-mos) and tissue plasminogen activator (tPA, Plat) mRNAs are dormant maternal mRNAs that are recruited during oocyte maturation; translation of Mos mRNA results in the activation of MAP kinase. **dsRNA** directed towards Mos or Plat mRNAs in mouse oocytes effectively results in the specific reduction of the targeted mRNA in both a time- and concentration-dependent manner. Moreover, **dsRNA** is more potent than either sense or **antisense** RNAs. Targeting the Mos mRNA results in inhibiting the appearance of MAP kinase activity and can result in parthenogenetic activation. Mos **dsRNA**, therefore, faithfully phenocopies the Mos null mutant. Targeting Plat mRNA with Plat **dsRNA** results in inhibiting production of tPA activity. Finally, effective reduction of the Mos and Plat mRNA is observed with stoichiometric amounts of Mos and Plat **dsRNA**, respectively.

L15 ANSWER 3 OF 69 CAPLUS COPYRIGHT 2001 ACS DUPLICATE 1
ACCESSION NUMBER: 2000:800864 CAPLUS
TITLE: Evidence that processed small dsRNAs may mediate sequence-specific mRNA degradation during RNAi in Drosophila embryos
AUTHOR(S): Yang, Dun; Lu, Hong; Erickson, James W.
CORPORATE SOURCE: Department of Biological Sciences, Columbia University, New York, NY, 10027, USA
SOURCE: Curr. Biol. (2000), 10(19), 1191-1200
CODEN: CUBLE2; ISSN: 0960-9822
PUBLISHER: Elsevier Science Ltd.
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Background: **RNA** interference (RNAi) is a phenomenon in which introduced **double-stranded** RNAs (dsRNAs) silence gene expression through specific degrdn. of their cognate mRNAs. Recent analyses in vitro suggest that dsRNAs may be copied, or converted, into 21-23 nucleotide (nt) guide RNAs that direct the nucleases responsible for RNAi to their homologous mRNA targets. Such small RNAs are also assocd. with gene silencing in plants. Results: We developed a quant. single-embryo assay to examine the mechanism of RNAi in vivo. We found that **dsRNA** rapidly induced mRNA degrdn. A fraction of dsRNAs were converted into 21-23 nt RNAs, and their time of appearance and persistence correlated precisely with **inhibition** of expression. The strength of RNAi increased disproportionately with increasing **dsRNA** length, but an 80 bp **dsRNA** was capable of effective gene silencing. RNAi was satd. at low **dsRNA** concn. and inhibited by excess unrelated **dsRNA**. The **antisense** strand of the **dsRNA** detd. target specificity, and excess complementary sense or **antisense** single-stranded RNAs (ssRNAs) competed with the RNAi reaction. Conclusions: Processed dsRNAs can act

directly to mediate RNAi, with the **antisense** strand detg. mRNA target specificity. The involvement of 21-23 nt RNAs is supported by the kinetics of the processing reaction and the obsd. size dependence. RNAi depends on a limiting factor, possibly the nuclease that generates the 21-23mer species. The active moiety appears to contain both sense and **antisense** RNA strands.


REFERENCE COUNT: 24
REFERENCE(S):
(1) Bass, B; Cell 2000, V101, P235 CAPLUS
(2) Bosher, J; Genetics 1999, V153, P1245 CAPLUS
(3) Bosher, J; Nat Cell Biol 2000, V2, PE31 CAPLUS
(4) Cogoni, C; Curr Opin Microbiol 1999, V2, P657 CAPLUS
(5) Cogoni, C; Nature 1999, V399, P166 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT


L15 ANSWER 4 OF 69 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.


ACCESSION NUMBER: 2000371589 EMBASE


TITLE: Molecular strategies for interrupting arthropod-borne virus transmission by mosquitoes.


AUTHOR: Blair C.D.; Adelman Z.N.; Olson K.E.
CORPORATE SOURCE: C.D. Blair, Department of Microbiology, Colorado State University, Fort Collins, CO 80523-1677, United States. cblair@cvmbs.colostate.edu



SOURCE: Clinical Microbiology Reviews, (2000) 13/4 (651-661). Refs: 97
ISSN: 0893-8512 CODEN: CMIREX
COUNTRY: United States
DOCUMENT TYPE: Journal; General Review
FILE SEGMENT: 004 Microbiology
037 Drug Literature Index
LANGUAGE: English
SUMMARY LANGUAGE: English



AB Arthropod-borne virus (arbovirus) infections cause a number of emerging and resurgent human and veterinary infectious diseases. Traditional means of controlling arbovirus diseases include vaccination of susceptible vertebrates and mosquito control, but in many cases these have been unavailable or ineffective, and so novel strategies for disease control are needed. One possibility is genetic manipulation of mosquito vectors to render them unable to transmit arboviruses. This review describes recent work to test the concept of pathogen-derived resistance in arthropods by expression of viral genes in mosquito cell cultures and mosquitoes. Sense and **antisense** genome sequences from La Crosse virus (LAC) (a member of the Bunyaviridae) and dengue viruses serotypes 1 to 4 (DEN-1 to DEN-4) (members of the Flaviviridae) were expressed in mosquito cells from **double**-subgenomic and replicon vectors based on Sindbis virus (a member of the Togaviridae). The cells were then challenged with homologous or related viruses. For LAC, expression of **antisense** sequences from the small (S) genome segment, particularly full-length **antisense** S **RNA**, effectively interfered with replication of challenge virus, whereas expression of either **antisense** or sense **RNA** from the medium (M) segment was completely ineffective in LAC **inhibition**. Expression of sense and **antisense** **RNA** derived from certain regions of the DEN genome also blocked homologous virus replication more effectively than did **RNA** from other regions. Other parameters of **RNA**-mediated interference have been defined, such as the time when replication is blocked and the minimum size of effector **RNA**. The mechanism of **RNA** **inhibition** has not been determined, although it resembles **double-stranded RNA** interference in other nonvertebrate systems. Prospects for application of molecular strategies to control arbovirus diseases are briefly reviewed.

L15 ANSWER 5 OF 69 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 2000:131900 CAPLUS
DOCUMENT NUMBER: 133:52982
TITLE: **Antisense** RNAs
AUTHOR(S): Branch, Andrea Denise
CORPORATE SOURCE: USA
SOURCE: Encycl. Microbiol. (2nd Ed.) (2000), Volume 1, 268-285. Editor(s): Lederberg, Joshua. Academic Press: San Diego, Calif.
CODEN: 68RKA9
DOCUMENT TYPE: Conference; General Review
LANGUAGE: English
AB A review with many refs. discussing **antisense** RNAs in prokaryotic systems (**inhibition** by direct binding to target RNAs), **antisense** RNAs in virus-infected mammalian cells (signals of danger), and artificial RNAs, **dsRNA**, and posttranscriptional gene silencing. (c) 2000 Academic Press.
REFERENCE COUNT: 32
REFERENCE(S): (2) Bass, B; Trends Biochem Sci 1997, V22, P157 CAPLUS
(3) Baulcombe, D; Plant Mol Biol 1996, V32, P79 CAPLUS
(4) Bram, R; Cell 1980, V19, P393 CAPLUS
(5) Branch, A; Trends Biochem Sci 1998, V23, P45 CAPLUS
(6) Delihas, N; Nat Biotechnol 1997, V15, P751 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L15 ANSWER 6 OF 69 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.
ACCESSION NUMBER: 2000280600 EMBASE
TITLE: **dsRNA**-mediated gene silencing in cultured Drosophila cells: A tissue culture model for the analysis of RNA interference.
AUTHOR: Caplen N.J.; Fleenor J.; Fire A.; Morgan R.A.
CORPORATE SOURCE: R.A. Morgan, Clinical Gene Therapy Branch, Natl. Human Genome Research Inst., National Institutes of Health, Bethesda, MD, United States. rmorgan@nhgri.nih.gov
SOURCE: Gene, (11 Jul 2000) 252/1-2 (95-105).
Refs: 36
ISSN: 0378-1119 CODEN: GENED6
PUBLISHER IDENT.: S 0378-1119(00)00224-9
COUNTRY: Netherlands
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 022 Human Genetics
LANGUAGE: English
SUMMARY LANGUAGE: English
AB **RNA** interference (RNAi) is a form of post-transcriptional gene silencing that has been described in a number of plant, nematode, protozoan, and invertebrate species. RNAi is characterized by a number of features: induction by **double stranded RNA** ( **dsRNA**), a high degree of specificity, remarkable potency and spread across cell boundaries, and a sustained down- regulation of the target gene. Previous studies of RNAi have examined this effect in whole organisms or in extracts thereof; we have now examined the induction of RNAi in tissue culture. A screen of mammalian cells from three different species showed no evidence for the specific down-regulation of gene expression by **dsRNA**. By contrast, RNAi was observed in Drosophila Schneider 2 (S2) cells. Green fluorescent protein (GFP) expression in S2 cells was inhibited in a dose-dependent manner by transfection of **dsRNA** corresponding to gfp when GFP was expressed either transiently or stably. This effect was structure- and sequence-specific in that: (1) little or no effect was seen when **antisense** (or sense) **RNA** was transfected; (2) an unrelated **dsRNA** did not reduce GFP expression; and (3) **dsRNA** corresponding to gfp had no effect on the expression of an unrelated target transgene. This invertebrate tissue culture model should

allow facile assays for loss of function in a well- defined cellular system and facilitate further understanding of the mechanism of RNAi and the genes involved in this process. (C) 2000 Elsevier Science B.V.

L15 ANSWER 7 OF 69 MEDLINE DUPLICATE 2
ACCESSION NUMBER: 2000117527 MEDLINE
DOCUMENT NUMBER: 20117527
TITLE: Distinct features of post-transcriptional gene silencing by **antisense** transgenes in single copy and inverted T-DNA repeat loci.
AUTHOR: Stam M; de Bruin R; van Blokland R; van der Hoorn R A; Mol J N; Kooter J M
CORPORATE SOURCE: Department of Developmental Genetics, Institute for Molecular Biological Sciences, BioCentrum Amsterdam, Vrije Universiteit, De Boelelaan 1087, 1081 HV Amsterdam, The Netherlands.
SOURCE: PLANT JOURNAL, (2000 Jan) 21 (1) 27-42.
Journal code: BRU. ISSN: 0960-7412.
PUB. COUNTRY: ENGLAND: United Kingdom
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200006
ENTRY WEEK: 20000604
AB The application of **antisense** transgenes in plants is a powerful tool to inhibit gene expression. The underlying mechanism of this **inhibition** is still poorly understood. High levels of **antisense RNA** (as-**RNA**) are expected to result in strong silencing but often there is no clear correlation between as-**RNA** levels and the degree of silencing. To obtain insight into these puzzling observations, we have analyzed several petunia transformants of which the pigmentation gene chalcone synthase (Chs) is post-transcriptionally silenced in corollas by **antisense** (as) Chs transgenes. The transformants were examined with respect to the steady-state as-**RNA** level, transcription level of the as-transgenes, the repetitiveness and structure of the integrated T-DNAs, and the methylation status of the transgenes. This revealed that the transformants can be divided in two classes: the first class contains a single copy (S) T-DNA of which the as-Chs gene is transcribed, although several-fold lower than the endogenous Chs genes. As there are not sufficient as-RNAs to degrade every mRNA, we speculate that silencing is induced by **double-stranded RNA**. The second class contains two T-DNAs which are arranged as inverted repeats (IRs). These IR loci are severely methylated and the as-Chs transgenes transcriptionally barely active. The strongest silencing was observed with IR loci in which the as-Chs transgenes were proximal to the centre of the IR. Similar features have been described for co-suppression by IRs composed of sense Chs transgenes, suggesting that silencing by **antisense** IRs also occurs by co-suppression, either via ectopic DNA pairing or via **dsRNA**.

L15 ANSWER 8 OF 69 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1999:819487 CAPLUS
DOCUMENT NUMBER: 132:60119
TITLE: **Inhibition** of the expression of BCR-ABL hybrid oncogene by **antisense** hairpin loop-RNA targeted to BCR-ABL fusion junction
INVENTOR(S): Stocks, Martin; Rabbitts, Terence
PATENT ASSIGNEE(S): Medical Research Council, UK
SOURCE: PCT Int. Appl., 49 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|---|---|---|---|---|
| WO 9967379 | A2 | 19991229 | WO 1999-GB1956 | 19990623 |
| WO 9967379 | A3 | 20000824 | | |

W: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM

RW: GH, GM, KE, LS, MW, SD, SL, SZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG

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| AU 9945197 | A1 | 20000110 | AU 1999-45197 | 19990623 |
| PRIORITY APPLN. INFO.: | | | GB 1998-13531 | 19980623 |
| | | | US 1998-90867 | 19980626 |
| | | | WO 1999-GB1956 | 19990623 |

AB A method of **inhibition** of the expression of BCR-ABL hybrid oncogene by **antisense** hairpin loop-RNA targeted to BCR-ABL fusion junction is described. The invention took advantage of the BCR-BCR fusion mRNA found in Philadelphia-pos. chronic myeloid leukemia (CML) and acute lymphocytic leukemia (ALL) resulting from the chromosome translocation t(9;20). To inhibit the expression of BCR-ABL hybrid oncogene expression, the **antisense** RNA targeted to the fusion junction of BCR-ABL hybrid mRNA was in vitro synthesized by T7 RNA polymerase from its vector or directly expressed in vivo from its expression vector. The **antisense RNA** was stabilized by spontaneous folding upon the synthesis into a **double** hairpin structure with a short stretch of single-**stranded** region between the two hairpins. The single stranded-region can initiate specific hybridization to the fusion junction of the target (p190 mRNA) with 7 bases complementary to 3' end of ABL moiety and 1 base complementary to 5' end of BCR moiety in BCR-ABL hybrid mRNA. The two hairpins will unwind and allow the full-length hybridization to the target mRNA (stabilized by much lower free energy than that required for the hairpin structure). The hairpin II at the 3' end of **antisense** RNA contained 31 bases complementary to the 5' sequence of BCR mRNA at the fusion junction and could be slightly different in order to differentiate the two isoforms of p190 mRNAs (.alpha. or .beta.). In the in vitro expt.,the **antisense** hAS190.alpha. only formed a hybrid to p190 RNA but not to p210 and ABL or BCR RNAs while the **antisense** hAS210 only hybridizes to p210 mRNA. The efficacy and specificity of these **antisense** RNAs were also demonstrated in vivo by cotransfecting COS7 or Hela cells with either preformed **antisense** RNA or its expression vector with its target gene expression vectors. The prodn. of target mRNA and protein was decreased significantly in the presence of the **antisense** RNA but the control mRNA and protein were not affected. The **antisense** mols. made by this strategy display increased specificity and stability of binding to target mRNA which can be used to control harmful expression in related disorders.

L15 ANSWER 9 OF 69 CAPLUS COPYRIGHT 2001 ACS

ACCESSION NUMBER: 1999:819486 CAPLUS

DOCUMENT NUMBER: 132:60098

TITLE: **Antisense** oligonucleotide constructs based on .beta.-D-arabinofuranose and its analogs

INVENTOR(S): Damha, Massad Jose; Parniak, Michael A.; Noronha, Anne M.; Wilds, Christopher; Borkow, Gadi; Arion, Dominique

PATENT ASSIGNEE(S): McGill University, Can.
SOURCE: PCT Int. Appl., 91 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|---|---|---|---|---|
| WO 9967378 | A1 | 19991229 | WO 1999-CA571 | 19990617 |
| W: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM | | | | |
| RW: GH, GM, KE, LS, MW, SD, SL, SZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG | | | | |
| AU 9945953 | A1 | 20000110 | AU 1999-45953 | 19990617 |
| PRIORITY APPLN. INFO.: | | | CA 1998-2241361 | 19980619 |
| | | | WO 1999-CA571 | 19990617 |

AB The present invention relates to modified oligonucleotide therapeutic agents to selectively prevent gene transcription and expression in a sequence-specific manner. In particular, this invention relates to the selective **inhibition** of protein biosynthesis via **antisense** strategy using oligonucleotides constructed from arabinonucleotide or modified arabinonucleotide residues. More particularly this invention relates to the use of **antisense** oligonucleotides having .beta.-D-arabinofuranose, 2-deoxy,2,2-difluoro-.beta.-D-ribose, or 2-deoxy-2-fluoro-.beta.-D-arabinose sugars to hybridize to complementary RNA such as cellular mRNA, viral RNA, etc. Arabinonucleoside oligomers serve as excellent models of **antisense** agents that have enhanced resistance to the action of degradative nucleases, bind to RNA through duplex formation, elicit RNase H activity, and inhibit in vitro and intracellular specific gene expression by binding to duplex DNA to form triple helixes. Accordingly, arabinonucleosides and its analogs have potential utility as therapeutic agents and/or tools for the study and control of specific gene expression in cells and organisms.

REFERENCE COUNT: 12
REFERENCE(S):
(1) Altmann, K; Antisense Oligonucleotide Technology 1998, P73 CAPLUS
(2) Aoyagi, M; Bioorganic & Medicinal Chemistry Letters 1996, V6, P1573 CAPLUS
(3) Damha, M; Journal of the American Chemical Society 1998, V120(49), P12976 CAPLUS
(5) Giannaris, P; Canadian Journal of Chemistry 1994, V72(3), P909 CAPLUS
(6) Gilead Sciences Inc; WO 9310820 A 1993 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L15 ANSWER 10 OF 69 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1999:425802 CAPLUS
DOCUMENT NUMBER: 131:54712
TITLE: **Inhibition** of gene expression via injection of **double-stranded RNA**
INVENTOR(S): Fire, Andrew; Xu, Siqun; Montgomery, Mary K.; Kostas, Stephen A.; Timmons, Lisa; Tabara, Hiroaki; Driver, Samuel E.; Mello, Craig C.
PATENT ASSIGNEE(S): The Carnegie Institute of Washington, USA
SOURCE: PCT Int. Appl., 54 pp.
CODEN: PIXXD2

DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|---|---|---|---|---|
| WO 9932619 | A1 | 19990701 | WO 1998-US27233 | 19981221 |
| W: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, US, US, UZ, VN, YU, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM | | | | |
| RW: GH, GM, KE, LS, MW, SD, SZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG | | | | |
| AU 9919380 | A1 | 19990712 | AU 1999-19380 | 19981221 |
| EP 1042462 | A1 | 20001011 | EP 1998-964202 | 19981221 |
| R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, FI | | | | |

PRIORITY APPLN. INFO.: US 1997-68562 19971223
US 1998-215257 19981218
WO 1998-US27233 19981221

AB The invention provides a process for introducing **RNA** into a living cell to inhibit expression of a target gene in that cell, whereby the **RNA** is **double-stranded RNA** ( **dsRNA**) and **inhibition** is sequence-specific in that the nucleotide sequences of the duplex region of the **RNA** and of a portion of the target gene are identical. The invention has been used to inhibit expression of 18 different genes from C. elegans, including unc-22, unc-54, fem-1, and hlh-1. **Antisense** interference, triple-strand interference, and co-suppression are known methods of gene **inhibition**, but the present invention offers advantages over these, including the ease of introducing **double-stranded RNA** (**dsRNA**) into cells, the low concn. of **RNA** which can be used, the stability of **dsRNA**, and the effectiveness of the **inhibition**. Unlike other methods, this invention does not suffer from being limited to a particular set of target genes, a particular portion of the target gene's nucleotide sequence, or a particular transgene or viral delivery method.

REFERENCE COUNT: 6
REFERENCE(S):
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(2) Fire, A; NATURE 1998, V391(6669), P806 CAPLUS
(3) Matzke, M; PLANT PHYSIOLOGY 1995, V107(3), P679 CAPLUS
(4) Montgomery, M; TRENDS IN GENETICS 1998, V14(7), P255 CAPLUS
(6) Timmons, L; NATURE 1998, V395(6705), P854 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L15 ANSWER 11 OF 69 MEDLINE DUPLICATE 3
ACCESSION NUMBER: 2000037831 MEDLINE
DOCUMENT NUMBER: 20037831
TITLE: A novel negative cis-regulatory element on the hepatitis B virus S-(+)-strand.
AUTHOR: Wagner M; Alt M; Hofschneider P H; Renner M
CORPORATE SOURCE: Department of Virus Research, Max-Planck-Institut fur Biochemie, Martinsried, Germany.
SOURCE: JOURNAL OF GENERAL VIROLOGY, (1999 Oct) 80 ( Pt 10) 2673-83.
Journal code: I9B. ISSN: 0022-1317.
PUB. COUNTRY: ENGLAND: United Kingdom
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Cancer Journals

ENTRY MONTH: 200002
ENTRY WEEK: 20000204
AB Hepatitis B virus (HBV) has a **double-stranded** DNA genome. The minus-strand contains coding regions for all known HBV proteins and most of the cis-regulatory elements. Little is known about transcription from the S-(+)-strand and its regulation. Thus, the presence of regulatory elements located on the S-(+)-strand was investigated by inserting nt 1038-1783 of HBV in both orientations between the human cytomegalovirus (HCMV) promoter and a luciferase gene. Transfection experiments revealed that the plasmid containing this HBV DNA fragment in an orientation allowing expression from the S-(+)-strand (**antisense**) led to **inhibition** of luciferase gene expression compared to the plasmid containing this sequence in an orientation that allows gene expression from the L-(-)-strand (sense). Deletion analyses delimit the sequence essential for the inhibitory effect to a 150 bp region that also carries part of the enhancerII/core promoter complex. However, the possible influence of this regulatory element has been excluded in various experiments. The repressing HBV sequence acts in an orientation- and position-dependent manner; no **inhibition** was observed when this DNA element was inserted upstream of the HCMV promoter or downstream of the luciferase gene. Northern blot analyses revealed reduced luciferase mRNA steady-state levels in cells transfected with constructs containing the essential HBV sequence in **antisense** orientation compared to plasmids containing this sequence in sense orientation. Since nuclear run-on experiments showed similar transcription initiation rates with these plasmids, the diminished luciferase mRNA steady-state levels must be due to altered stabilities, suggesting that nt 1783-1638 of HBV encode an **RNA**-destabilizing element.

L15 ANSWER 12 OF 69 MEDLINE DUPLICATE 4
ACCESSION NUMBER: 1999197090 MEDLINE
DOCUMENT NUMBER: 99197090
TITLE: Growth **inhibition** by a triple ribozyme targeted to repetitive B2 transcripts.
AUTHOR: Crone T M; Schalles S L; Benedict C M; Pan W; Ren L; Loy S E; Isom H; Clawson G A
CORPORATE SOURCE: Departments of Pathology, The Cell and Molecular Biology Program, The Pennsylvania State University, Milton S. Hershey Medical Center, Hershey, PA, USA.
CONTRACT NUMBER: CA21141 (NCI)
CA40145 (NCI)
CA23931 (NCI)
SOURCE: HEPATOLOGY, (1999 Apr) 29 (4) 1114-23.
Journal code: GBZ. ISSN: 0270-9139.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199907
ENTRY WEEK: 19990704
AB The B2 family represents a group of short repetitive sequences that are found throughout the rodent genome and are analogous to the human Alu sequences. Certain B2 subfamilies are transcribed by **RNA** polymerase III (pol III), and this transcription is in part controlled by the retinoblastoma protein. In addition to their putative role in retrotranspositional events, these actively transcribed B2 RNAs show a predicted highly stable secondary structure. Although B2 transcripts are normally confined to the nucleus, they demonstrate altered compartmentation after carcinogen treatment, in cancers, and in immortalized and/or transformed cell lines, the significance of which is unclear. Because modulation of B2 transcripts did not seem feasible with an **antisense** approach, we designed a triple ribozyme (TRz)

construct to down-regulate B2 transcripts. The B2-targeted TRz undergoes efficient self-cleavage, resulting in liberation of the internal hammerhead Rz, which we targeted to a single-**stranded** region of the consensus B2 sequence. The liberated internal targeted Rz was 20 times more active than the corresponding **double**-G mutant construct that could not undergo self-cleavage, and 5 times more active than the same Rz flanked by nonspecific vector sequences. The B2-targeted TRz was used to develop stable transfectant clones from an SV40-immortalized hepatocyte cell line. These transfectant clones all showed variably reduced growth rates, accompanied by significant reductions in both cytoplasmic and nuclear B2 **RNA** levels: linear regression analyses showed that their growth rates were directly related to residual cytoplasmic B2 levels. Reverse-transcription polymerase chain reaction (RT-PCR) analyses documented efficient self-liberation of the internal targeted Rz in vivo, and showed that the relative cytoplasmic expression levels generally paralleled the magnitude of the decrease in B2 transcripts. The RT-PCR analyses further demonstrated that up to 20% of the Rz was located in the nucleus, which presumably reflects competition between autocatalytic processing and nucleocytoplasmic transport of the initial TRz transcript.

L15 ANSWER 13 OF 69 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1998:795126 CAPLUS
DOCUMENT NUMBER: 130:48296
TITLE: Cell growth-controlling **antisense** oligonucleotides which inhibit protein kinase R
INVENTOR(S): Petryshyn, Raymond A.
PATENT ASSIGNEE(S): Research Corporation Technologies, Inc., USA
SOURCE: PCT Int. Appl., 121 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|---|---|---|---|---|
| WO 9854315 | A1 | 19981203 | WO 1998-US10001 | 19980515 |
| W: CA, JP | | | | |
| RW: AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE | | | | |
| US 6124091 | A | 20000926 | US 1997-867230 | 19970530 |
| PRIORITY APPLN. INFO.: | | | US 1997-867230 | 19970530 |

AB Claimed are **antisense** oligonucleotides which inhibit a protein kinase R- (PKR-)activating protein by binding the portion of the RNA which would otherwise bind and stimulate autophosphorylation of PKR, thereby stimulating cell growth, and the cDNA sequence from which these oligonucleotides are derived. The present invention provides a partial cDNA corresponding to an **RNA** contg. **double stranded** regions (R-**RNA**), which, when transcribed in vitro, gives rise to an **RNA** transcript that activates PKR. An approx. 226-252 bp nucleotide (nt) sequence responsible for activation of PKR (the activation sequence) has been identified within the cDNA and isolated. **Antisense** oligonucleotides corresponding to specific portions of the 252 nt cDNA fragment stimulate proliferation of different cells in culture. Various portions of the cDNA or R-RNA may also be used to inhibit cell proliferation in cell cultures. The present invention further provides pharmaceutical compns. comprising the subject nucleic acid fragments and oligonucleotides. Kits which comprise at least one of the subject isolated nucleic acid mols. or oligonucleotides and a pharmaceutically acceptable carrier are also provided.

REFERENCE COUNT: 12
REFERENCE(S): (1) Anon; DATABASE STRAND 1996
(2) Anon; DATABASE STRAND 1996

(4) Khan, A; NATURE GENET 1992, V2, P180 CAPLUS
(5) Maitra, R; J BIOL CHEM 1995, V270, P15071 CAPLUS
(11) Petryshyn, R; NUCLEIC ACIDS RES 1997, V25(13), P2672 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L15 ANSWER 14 OF 69 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1998:112448 CAPLUS
DOCUMENT NUMBER: 128:176950
TITLE: Using **double-stranded RNA** -specific ribonucleases to increase the effectiveness of **antisense RNA** **inhibition** of gene expression
INVENTOR(S): Werner, Dieter; Granzow, Christof; Schubert, Marie; Rothbarth, Karsten; Dittmar, Gunnar; Stammer, Herrmann; Todorov, Ivan
PATENT ASSIGNEE(S): Deutsches Krebsforschungszentrum Stiftung des Offentlichen Rechts, Germany; Werner, Dieter; Granzow, Christof; Schubert, Marie; Rothbarth, Karsten; Dittmar, Gunnar; Stammer, Herrmann; Todorov, Ivan
SOURCE: PCT Int. Appl., 18 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: German
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|---|---|---|---|---|
| WO 9805771 | A1 | 19980212 | WO 1997-DE1692 | 19970805 |
| W: JP, US | | | | |
| RW: AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE | | | | |
| DE 19631918 | A1 | 19980212 | DE 1996-19631918 | 19960807 |

PRIORITY APPLN. INFO.: DE 1996-19631918 19960807
AB A method of increasing the effectiveness of **inhibition** of gene expression by **antisense RNA** in cells by expression of a gene for a **double-stranded RNA** specific nuclease (dsRNase) in the cells contg. the **antisense RNA** is described. The invention also concerns cells which express a (ds)RNAase and a combination of an **antisense** RNA and a (ds)RNAase which are coded by one or several vectors. Cells expressing genes for a dsRNase and and **antisense** RNA are described. The effectiveness of the method is demonstrated using a chloramphenicol acetyltransferase (CAT) reporter gene, the pac1+ gene of Schizosaccharomyces pombe encoding a dsRNase and an **antisense** gene for CAT in Ehrlich ascites cells.

L15 ANSWER 15 OF 69 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1998:112447 CAPLUS
DOCUMENT NUMBER: 128:177409
TITLE: **Antisense** RNAs with unusual secondary structures and the **inhibition** of gene expression
INVENTOR(S): Werner, Dieter; Granzow, Christof; Joswig, Gaby; Rothbarth, Karsten; Schubert, Marie
PATENT ASSIGNEE(S): Deutches Krebsforschungszentrum Stiftung des Offentlichen Rechts, Germany; Werner, Dieter; Granzow, Christof; Joswig, Gaby; Rothbarth, Karsten; Schubert, Marie
SOURCE: PCT Int. Appl., 14 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: German

FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|---|---|---|---|---|
| WO 9805770 | A2 | 19980212 | WO 1997-DE1691 | 19970805 |
| WO 9805770 | A3 | 19980326 | | |

W: JP, US
RW: AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE

| | | | | |
|---|---|---|---|---|
| DE 19631919 | A1 | 19980212 | DE 1996-19631919 | 19960807 |
| DE 19631919 | C2 | 19980716 | | |
| EP 918853 | A2 | 19990602 | EP 1997-936610 | 19970805 |

R: AT, BE, CH, DE, DK, ES, FR, GB, IT, LI, NL, SE

PRIORITY APPLN. INFO.: DE 1996-19631919 19960807
WO 1997-DE1691 19970805

AB The **antisense** RNA and its combination may be used to inhibit gene expression. **Antisense** RNAs that have an unusual long **double-stranded** region with a hairpin loop are described for use in the **inhibition** of gene expression, either on its own or in combination with a **double-stranded RNA** -specific RNase. The sequence (GC)20-GAATTC-(GC)20 is used to create the double-stranded region. The GAATTC sequence can be replaced with any short palindromic sequence.

L15 ANSWER 16 OF 69 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1998:774162 CAPLUS
DOCUMENT NUMBER: 130:24138
TITLE: Increasing the efficiency of manufacture of viral antigens for vaccines by **inhibition** of **double-stranded RNA** -activated protein kinase synthesis
INVENTOR(S): Lau, Allan S.
PATENT ASSIGNEE(S): The Regents of the University of California, USA
SOURCE: U.S., 12 pp.
CODEN: USXXAM
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|---|---|---|---|---|
| US 5840565 | A | 19981124 | US 1996-700198 | 19960821 |

AB Methods for increasing yields of viral antigens by replication of a vaccine strain of a virus in animal cell culture are described. These methods rely on the manipulation of the cellular levels of certain interferon induced antiviral activities, in particular, cellular levels of **double-stranded RNA** (**dsRNA**) dependent kinase (PKR). PKR-deficient cells are obtained by any one of four methods. Cells can be transformed with an **antisense** DNA or they may be allowed to passively uptake the **antisense** DNA. Cells may be transformed with a dominant neg. mutant of the kinase gene. Cells can be cultured in the presence of 2-aminopurine. In cell cultures deficient for PKR, virus yield is increased by several orders of magnitude over cell cultures with normal levels of these proteins making these cell cultures useful for the prodn. of viral vaccines.

REFERENCE COUNT: 55
REFERENCE(S):
(2) Barber; Proc Natl Acad Sci USA 1994, V91, P4278 CAPLUS
(3) Bowie, J; Science 1990, V247(4948), P1306 CAPLUS
(5) Busby; J Mol Biol 1982, V154, P197 CAPLUS
(6) Camper; Biology of Reproduction 1995, V52, P246 CAPLUS

L15 ANSWER 17 OF 69 MEDLINE DUPLICATE 5
ACCESSION NUMBER: 1998411319 MEDLINE
DOCUMENT NUMBER: 98411319
TITLE: Tumor suppressor p53 as a component of the tumor necrosis factor-induced, protein kinase PKR-mediated apoptotic pathway in human promonocytic U937 cells.
AUTHOR: Yeung M C; Lau A S
CORPORATE SOURCE: The Moses Grossman Infectious Diseases Laboratory, Department of Pediatrics, San Francisco General Hospital and University of California, San Francisco, California 94110, USA.
SOURCE: JOURNAL OF BIOLOGICAL CHEMISTRY, (1998 Sep 25) 273 (39) 25198-202.
Journal code: HIV. ISSN: 0021-9258.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Cancer Journals
ENTRY MONTH: 199812
ENTRY WEEK: 19981203
AB Despite what is known about the early signaling events in tumor necrosis factor (TNF) alpha-induced apoptosis, characterization of the downstream events remains largely undefined. It is now known that a cross-talk exists between the interferon and TNF-alpha pathways. This linkage allows recruitment of the cell proliferation suppressor PKR (**dsRNA** -dependent protein kinase) from the interferon pathway to play a pivotal role in TNF-alpha-induced apoptosis. In this study, we took advantage of the differential TNF-alpha susceptibilities of human promonocytic U937 subclones, deficient in or overexpressing PKR, to further characterize the role of PKR in apoptosis. By reverse transcription-polymerase chain reaction, we demonstrated that TNF-alpha transiently induces the tumor suppressor p53 in U937 cells. This p53 induction lags behind the TNF-alpha induction of PKR by 1 h. By cell viability determination, ultrastructural studies, apoptotic DNA laddering, and **antisense** techniques, it was shown that **inhibition** of p53 expression in PKR-overexpressing U937 cells abrogates the TNF-alpha-induced apoptosis in these cells. Conversely, overexpressing wild type p53 in PKR-deficient U937 cells confers the susceptibility of these cells to TNF-alpha-induced apoptosis. This latter result indicates that p53 induction is an event downstream of TNF-alpha-induced up-regulation of PKR, thereby further establishing the critical role of p53 in TNF-alpha-induced apoptosis in U937 cells. PKR-overexpressing U937 cells were found to possess a constitutively higher level of p53, which partly explains why these cells spontaneously undergo apoptosis even without TNF-alpha treatment. Finally, a model is presented on the interplay between PKR and p53 in effecting TNF-alpha-induced apoptosis in U937 cells.

L15 ANSWER 18 OF 69 MEDLINE DUPLICATE 6
ACCESSION NUMBER: 1998248568 MEDLINE
DOCUMENT NUMBER: 98248568
TITLE: Selective **inhibition** of cell-free translation by oligonucleotides targeted to a mRNA hairpin structure.
AUTHOR: Le Tinevez R; Mishra R K; Toulme J J
CORPORATE SOURCE: INSERM U 386, IFR Pathologies Infectieuses, Universite Victor Segalen, 146 rue Leo Saignat, 33076 Bordeaux cedex, France.
SOURCE: NUCLEIC ACIDS RESEARCH, (1998 May 15) 26 (10) 2273-8.
Journal code: O8L. ISSN: 0305-1048.

PUB. COUNTRY: ENGLAND: United Kingdom
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Cancer Journals
ENTRY MONTH: 199809
ENTRY WEEK: 19980902

AB Using an in vitro selection approach we have previously isolated oligodeoxy aptamers that can bind to a DNA hairpin structure without disrupting the **double-stranded** stem. We report here that these oligomers can bind to the **RNA** version of this hairpin, mostly through pairing with a designed 6 nt anchor. The part of the aptamer selected against the DNA hairpin did not increase stability of the **RNA**-aptamer complex. However, it contributed to the binding site for Escherichia coli RNase H, leading to very efficient cleavage of the target **RNA**. In addition, a 2'- O -methyloligoribonucleotide analogue of one selected sequence selectively blocked in vitro translation of luciferase in wheat germ extract by binding to the hairpin region inserted upstream of the initiation codon of the reporter gene. Therefore, non-complementary oligomers can exhibit **antisense** properties following hybridization with the target **RNA**. Our study also suggests that in vitro selection might provide a means to extend the repertoire of sequences that can be targetted by **antisense** oligonucleotides to structured **RNA** motifs of biological importance.

L15 ANSWER 19 OF 69 LIFESCI COPYRIGHT 2001 CSA
ACCESSION NUMBER: 1998:26158 LIFESCI
TITLE: **Double-stranded RNA** poses puzzle
AUTHOR: Wagner, R.W.; Sun, Lin
CORPORATE SOURCE: Phylos Inc., 300 Putnam Ave., Cambridge, MA 02139, USA
SOURCE: NATURE, (19980200) vol. 391, no. 6669, pp. 744-745. ISSN: 0028-0836.
DOCUMENT TYPE: Journal
TREATMENT CODE: General Review
FILE SEGMENT: N
LANGUAGE: English

AB The human genome is predicted to contain between 50,000 and 100,000 genes. To work out what these genes do, an array of techniques is needed to evaluate the protein-protein interactions and biochemical pathway of any gene product. The nematode worm Caenorhabditis elegans is an excellent system for such studies because of its well-understood genetics and development, evolutionary conservation to human genes, small genome size and relatively short life cycle. The 100-megabase-pair genome will be completely sequenced this year, and a total of 17,000 genes have been predicted, many with human counterparts. Approaches used to manipulate gene expression in C. elegans include transposon-mediated deletion, **antisense inhibition** and direct isolation of deletions after mutagenesis. Although these methods have proved useful, limitations still exist. On page 806 of this issue, Fire and colleagues describe a remarkable and surprising technique for inhibiting gene function in C. elegans. They turned off a specific gene in progeny worms by microinjecting **double-stranded RNA** ( **dsRNA**) complementary to the coding region of the gene into the gonads of adult animals. Using a well-characterized gene, unc-22, which encodes a non-essential myofilament protein, they showed that injection of **dsRNA** produced a phenotype characteristic of unc-22 **inhibition**--twitching.

L15 ANSWER 20 OF 69 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1998:34900 CAPLUS

DOCUMENT NUMBER: 128:164062
TITLE: Theoretical design of **antisense** RNA structures substantially improves annealing kinetics and efficacy in human cells
AUTHOR(S): Patzel, Volker; Sczakiel, Georg
CORPORATE SOURCE: Forschungsschwerpunkt Angewandte Tumorvirologie, Deutsches Krebsforschungszentrum, Heidelberg, D-69120, Germany
SOURCE: Nat. Biotechnol. (1998), 16(1), 64-68
CODEN: NABIF9; ISSN: 1087-0156
PUBLISHER: Nature America
DOCUMENT TYPE: Journal
LANGUAGE: English

AB The success of **antisense** therapeutics is not predictable despite their widespread use in biotechnol. and mol. medicine. The relationship between RNA structure and biol. effectiveness is largely not understood; however, **antisense** RNA-mediated effects in vivo seem to be related to annealing kinetics in vitro. This study suggests that terminal unpaired nucleotides and overall flexibility of **antisense** RNA directed against the human immunodeficiency virus type 1 (HIV-1) are related to fast RNA-RNA annealing in vitro as well as to strong **inhibition** of virus replication in human cells. Annealing rate consts. of computer-selected **antisense** RNA species approach the values for natural **antisense** RNA in the order of 106 M-1s-1. When considering the unfavorable stability in cellular exts. of **antisense** RNA species that were found to anneal fast in vitro, an **antisense** effect against HIV-1 in human cells was obsd. that was 10- to 10,000-fold stronger than that measured for species predicted to anneal slowly. A computer-supported structural design of **antisense** RNA can serve as a platform to det. RNA-RNA assocn. in vitro and biol. effectiveness in living cells.

=> d l15 ibib abs 21-30

L15 ANSWER 21 OF 69 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1997:684493 CAPLUS
DOCUMENT NUMBER: 127:355924
TITLE: Sets of **antisense** oligonucleotides with increased specificity that form partially double-stranded hybrids
INVENTOR(S): Kandimalla, Ekambar R.; Agrawal, Sudhir
PATENT ASSIGNEE(S): Hybridon, Inc., USA
SOURCE: PCT Int. Appl., 91 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|---|---|---|---|---|
| WO 9738097 | A1 | 19971016 | WO 1997-US5683 | 19970404 |
| W: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, FI, GB, GE, HU, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, TJ, TM, TR, TT, UA, UG, UZ, VN, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM | | | | |
| RW: GH, KE, LS, MW, SD, SZ, UG, AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG | | | | |
| AU 9727234 | A1 | 19971029 | AU 1997-27234 | 19970404 |
| PRIORITY APPLN. INFO.: | | | US 1996-627967 | 19960404 |

AB A method of increasing the specificity of **antisense** oligonucleotides without increasing their length is described. The method uses a set of oligonucleotides capable of hybridizing to target sequences within a a few base pairs of one another. The oligonucleotides have a domain that binds to the target sequence and a domain that allows it to bind to the other oligonucleotide of the pair. The partially double stranded oligonucleotides has two specific binding domains that give greater specificity and stability than a single domain of comparable size.

Two or more such oligonucleotides can be used together. These oligonucleotides can be used in the therapeutic **inhibition** of gene expression, e.g. in the treatment of viral infection. Optimization expts. in which oligonucleotides designed to inhibit transcription of the gag gene of HIV-1 were developed and characterized are reported.

L15 ANSWER 22 OF 69 MEDLINE DUPLICATE 7
ACCESSION NUMBER: 1998052529 MEDLINE
DOCUMENT NUMBER: 98052529
TITLE: **Inhibition** of interferon regulatory factor-1 expression results in predominance of cell growth stimulatory effects of interferon-gamma due to phosphorylation of Stat1 and Stat3.
AUTHOR: Sato T; Selleri C; Young N S; Maciejewski J P
CORPORATE SOURCE: Hematology Branch, National Heart, Lung, and Blood Institute, Bethesda, MD, USA.
SOURCE: BLOOD, (1997 Dec 15) 90 (12) 4749-58.
Journal code: A8G. ISSN: 0006-4971.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals; Cancer Journals
ENTRY MONTH: 199803
ENTRY WEEK: 19980301

AB Interferon-gamma (IFN-gamma) is a potent inhibitor of hematopoiesis in vitro and has been implicated in the pathophysiology of human bone marrow failure syndromes. IFN-gamma both inhibits cell cycling and induces expression of the Fas-receptor,, resulting in subsequent apoptosis of hematopoietic progenitor cells. IFN regulatory factor-1 (IRF-1) mediates some of these suppressive effects by activation of downstream inducible genes, such as **double-stranded RNA** -activatable protein kinase and inducible nitric oxide synthase. However, under certain experimental conditions, IFN-gamma appears to stimulate proliferation of hematopoietic cells. Based on the hypothesis that IFN-gamma-receptor triggering may activate diverse signaling cascades, we designed experiments to determine which intracellular mechanisms (in addition to the IRF-1 transduction pathway) influence the biologic effects of IFN-gamma. Using **antisense** technique, we inhibited the IRF-1-mediated pathway in KG1a cells stimulated with IFN-gamma. In contrast to the suppressive effects of IFN-gamma observed in control cells, untreated and IFN-gamma-treated KG-1a cells that were transduced with retroviral vectors expressing IRF-1 **antisense** mRNA showed enhanced proliferation. The increased growth rate was associated with decreased levels of IRF-1 mRNA and protein but unchanged levels of IRF-2. We inferred that IFN-gamma could also activate a stimulatory transduction pathway that, under specific conditions, may control the cellular response to this cytokine. The family of Stat proteins is involved in signal transduction of hematopoietic growth factors. We showed that, in KG-1a cells, IFN-gamma also induced phosphorylation of Stat1 and Stat3, whereas p42 MAP kinase was phosphorylated regardless of the presence of IFN-gamma.

Using electrophoresis mobility shift assays, IFN-gamma enhanced

Stat1-Stat1 homodimer and Stat1-Stat3 heterodimer formation, suggesting that, in addition to inhibitory signals mediated by IRF-1, IFN-gamma may activate proliferative signals by phosphorylation of Stat1 and Stat3 proteins. The observations made in experiments with KG-1a cells were confirmed in primary hematopoietic cells. After **inhibition** of the IRF-1 pathway by transduction of an **antisense** IRF-1 retrovirus into human CD34+ cells, IFN-gamma produced an aberrant stimulatory effect on hematopoietic colony formation. Conversely, in control vector-transduced CD34+ cells, the typical inhibitory response to IFN-gamma was seen. Our results indicate that inhibitory cytokines such as IFN-gamma may exhibit diverse biologic effects depending on the intracellular balance of transcriptional regulators, in turn influenced by the activation and differentiation status of the target cells.

L15 ANSWER 23 OF 69 MEDLINE DUPLICATE 8
ACCESSION NUMBER: 97388552 MEDLINE
DOCUMENT NUMBER: 97388552
TITLE: 2',5'-linked oligo-3'-deoxyribonucleoside phosphorothioate chimeras: thermal stability and **antisense inhibition** of gene expression.
AUTHOR: Bhan P; Bhan A; Hong M; Hartwell J G; Saunders J M; Hoke G D
CORPORATE SOURCE: Dyad Pharmaceutical Corporation, 9110 Red Branch Road, Columbia, MD 21045, USA.. purshotam.bhan@am.pharmacia.com
CONTRACT NUMBER: R44 GM49581-02 (NIGMS)
SOURCE: NUCLEIC ACIDS RESEARCH, (1997 Aug 15) 25 (16) 3310-7. Journal code: O8L. ISSN: 0305-1048.
PUB. COUNTRY: ENGLAND: United Kingdom
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Cancer Journals
ENTRY MONTH: 199711
ENTRY WEEK: 19971103
AB 2',5'-Linked oligo-3'-deoxyribonucleotides bind selectively to complementary **RNA** but not to DNA. These oligonucleotides (ODNs) do not recognize **double-stranded** DNA by Hoogsteen triplex formation and the complexes formed by these ODNs with **RNA** are not substrates for Escherichia coli RNase H. Substitution of the 2',5'-phosphodiester backbone by phosphorothioate linkages gives 2',5'-linked oligo-3'-deoxynucleoside phosphorothioate ODNs that exhibit significantly less non-specific binding to cellular proteins or thrombin. Incorporation of a stretch of seven contiguous 3',5'-linked oligo-2'-deoxynucleoside phosphorothioate linkages in the center of 2',5'-linked ODNs (as a putative RNase H recognition site) afford chimeric **antisense** ODNs that retain the ability to inhibit steroid 5alpha-reductase (5alphaR) expression in cell culture.

L15 ANSWER 24 OF 69 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1997:96770 CAPLUS
DOCUMENT NUMBER: 126:86393
TITLE: Binding Affinity and Specificity of Escherichia coli RNase H1: Impact on the Kinetics of Catalysis of **Antisense** Oligonucleotide-RNA Hybrids
AUTHOR(S): Lima, Walt F.; Crooke, Stanley T.
CORPORATE SOURCE: Isis Pharmaceuticals Inc., Karlovy vary, CA, 92008, USA
SOURCE: Biochemistry (1997), 36(2), 390-398
CODEN: BICHAW; ISSN: 0006-2960
PUBLISHER: American Chemical Society
DOCUMENT TYPE: Journal
LANGUAGE: English
AB In this study we report for the first time the binding affinity of RNase H1 for oligonucleotide duplexes. We used a previously described 17-mer

**antisense** sequence [Monia, B. P., Johnston, J. F., Ecker, D. J., Zounes, M. A., Lima, W. F., & Freier, S. M. (1992) J. Biol. Chem. 267, 19954-19962] hybridized to a complementary oligoribonucleotide to evaluate both the binding affinity and the catalytic rate of RNase H1. The dissocn. consts. (Kd) of RNase H1 for the various substrates tested were detd. by **inhibition** anal. using chem. modified noncleavable oligonucleotide heteroduplexes. Catalytic rates were detd. using heteroduplex substrates contg. chimeric **antisense** oligonucleotides composed of a five-base deoxynucleotide sequence flanked on either side by chem. modified nucleotides. We find that the enzyme preferentially binds A-form duplexes: RNase H bound A-form duplexes (RNA:RNA and DNA:RNA) approx. 60-fold tighter than B-form duplexes (DNA:DNA) and approx. 300-fold tighter than single-strand oligonucleotides. The enzyme exhibited equal affinity for both the wild type (RNA:DNA) oligonucleotide substrate and heteroduplexes contg. various 2'-sugar modifications, while the cleavage rates for these chem. modified substrates were without exception slower than for the wild type substrate.

The introduction of a single pos. charged 2'-propoxyamine modification into the chimeric **antisense** oligonucleotide portion of the heteroduplex substrate resulted in both decreased binding affinity and a slower rate of catalysis by RNase H. The cleavage rates for heteroduplexes contg. single-base mismatch sequences within the chimeric oligonucleotide portion varied depending on the position of the mismatch but had no effect on the binding affinity of the enzyme. These results offer further insights into the phys. binding properties of the RNase H-substrate interaction as well as the design of effective **antisense** oligonucleotides.

L15 ANSWER 25 OF 69 MEDLINE

ACCESSION NUMBER: 1998247171 MEDLINE
DOCUMENT NUMBER: 98247171
TITLE: **Inhibition** of HIV-1 replication by foldback triple-helix forming oligonucleotides.
AUTHOR: Hiratou T; Tsukahara S; Takai K; Koyanagi Y; Yamamoto N; Takaku H
CORPORATE SOURCE: Department of Industrial Chemistry, Chiba Institute of Technology, Japan.
SOURCE: NUCLEIC ACIDS SYMPOSIUM SERIES, (1997) (37) 221-2. Journal code: O8N. ISSN: 0261-3166.
PUB. COUNTRY: ENGLAND: United Kingdom
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199809
ENTRY WEEK: 19980904

AB Replication of retroviral **RNA** into **double-stranded** DNA is catalyzed by reverse transcriptase (RT). The polypurine tract (PPT) serves as a primer for plus-strand DNA synthesis and is highly conserved among HIV-1. The PPT region is a possible target for triple-helix formation. Here, we show the effects of triple-helix formation by analyses of melting temperature and gel shift using a foldback triplex-forming-oligonucleotides (FTFOs). We found that the FTFOs containing phosphorothioate groups at the 3'- and 5'-ends, or inside the hairpin loop, exhibited greater exonuclease resistance than the unmodified FTFOs. Several triplex oligonucleotides have thermal stability. The abilities of the FTFOs (DsDG-37) containing the guanosine in place of the cytidine in the third Hoogsteen base-pairing strand to inhibit HIV-1 replications were examined. The FTFOs (DsDG-37) inhibit the replication of HIV-1 more efficiently than the FTFOs (DsD-37) indicating sequence-specific **inhibition** of HIV-1 replication.

L15 ANSWER 26 OF 69 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.
ACCESSION NUMBER: 96335832 EMBASE
DOCUMENT NUMBER: 1996335832
TITLE: An essential role for the interferon-inducible, **double-stranded RNA**- activated protein kinase PKR in the tumor necrosis factor-induced apoptosis in U937 cells.
AUTHOR: Yeung M.C.; Liu J.; Lau A.S.
CORPORATE SOURCE: San Francisco General Hospital, 1001 Potrero Avenue,San Francisco, CA 94110, United States
SOURCE: Proceedings of the National Academy of Sciences of the United States of America, (1996) 93/22 (12451-12455). ISSN: 0027-8424 CODEN: PNASA6
COUNTRY: United States
DOCUMENT TYPE: Journal; Conference Article
FILE SEGMENT: 026 Immunology, Serology and Transplantation
029 Clinical Biochemistry
037 Drug Literature Index
LANGUAGE: English
SUMMARY LANGUAGE: English
AB Tumor necrosis factor .alpha. (TNF-.alpha.) is well-characterized for its necrotic action against tumor cells; however, it has been increasingly associated with an apoptosis-inducing potential on target cells. While the signaling events and the actual cytolytic mechanism(s) for both TNF-.alpha.-induced necrosis and apoptosis remain to be fully elucidated, we report here on (i) the ability of TNF-.alpha. to induce apoptosis in the promonocytic U937 cells, (ii) the discovery of a cross-talk between the TNF-.alpha. and the interferon signaling pathways, and (iii) the pivotal role of interferon-inducible, **double-stranded RNA**-activated protein kinase (PKR) in the induction of apoptosis by TNF-.alpha.. Our data from microscopy studies, trypan blue exclusion staining, and apoptotic DNA ladder electrophoresis revealed that a subclone derived from U937 and carrying a PKR **antisense** expression vector was resistant to TNF-.alpha.-induced apoptosis. Further, TNF-.alpha. initiated a generalized **RNA** degradation process in which the participation of PKR was required. Finally, the PKR gene is a candidate 'death gene' since overexpression of this gene could bring about apoptosis in U937 cells.

L15 ANSWER 27 OF 69 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1996:683903 CAPLUS
DOCUMENT NUMBER: 126:436
TITLE: **Inhibition** of HIV-1 replication by oligonucleotides forming triple-helixes targeted to polypurine tract
AUTHOR(S): Tsukahara, Satoru; Suzuki, Junji; Goto, Yuta; Inagawa, Takubumi; Takeuchi, Hiroaki; Takai, Kazuyuki; Koyanagi, Yoshio; Yamamoto, Naoki; Takaku, Hiroshi
CORPORATE SOURCE: Dep. Industrial Chem., Chiba Inst. Technol., Chiba, 275, Peop. Rep. China
SOURCE: Nucleic Acids Symp. Ser. (1996), 35(Twentythird Symposium on Nucleic Acids Chemistry, 1996), 181-182 CODEN: NACSD8; ISSN: 0261-3166
PUBLISHER: Oxford University Press
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Replication of retroviral **RNA** into **double-stranded** DNA is catalyzed by reverse transcriptase (RT). The polypurine tract (PPT) serves as a primer for plus-strand DNA synthesis and is highly conserved among HIV-1. The PPT region is a possible target for triple-helix formation. Here, we show the effects of triple-helix

formation by analyses of melting temp. and protection from reverse transcription in vitro using two systems (two-strand or three-strand-system). Furthermore, we used phosphorothioate oligonucleotide probes to increase the nuclease resistance. Several triplex oligonucleotides have thermal stability and prevent the initiation of minus-strand DNA synthesis by RT. We also demonstrate **inhibition** of HIV-1 replication by these oligonucleotides.

L15 ANSWER 28 OF 69 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.
ACCESSION NUMBER: 96036003 EMBASE
DOCUMENT NUMBER: 1996036003
TITLE: **Antisense** strategies and therapeutic applications.
AUTHOR: Putnam D.A.
CORPORATE SOURCE: Controlled Chemical Delivery Center, College of Pharmacy, University of Utah,Salt Lake City, UT 84112, United States
SOURCE: American Journal of Health-System Pharmacy, (1996) 53/2 (151-160).
ISSN: 1079-2082 CODEN: AHSPEK
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 022 Human Genetics
027 Biophysics, Bioengineering and Medical Instrumentation
029 Clinical Biochemistry
030 Pharmacology
037 Drug Literature Index
LANGUAGE: English
SUMMARY LANGUAGE: English

AB The concepts underlying the **antisense** approach to disease therapy are discussed, and potential applications are examined. **Antisense** therapeutic agents bind to DNA or **RNA** sequences, blocking the synthesis of cellular proteins with unparalleled specificity. Transcription and translation are the two processes with which the agents interfere. There are three major classes of **antisense** agents: **antisense** sequences, commonly called **antisense** oligonucleotides; antigene sequences; and ribozymes. **Antisense** sequences are derivatives of nucleic acids that hybridize cytosolic messenger **RNA** (mRNA) sense strands through hydrogen bonding to complementary nucleic acid bases. Antigene sequences hybridize **double-stranded** DNA in the nucleus, forming triple helixes. Ribozymes, rather than inhibiting protein synthesis simply by binding to a single targeted mRNA, combine enzymatic processes with the specificity of **antisense** base pairing, creating a molecule that can incapacitate multiple targeted mRNAs. **Antisense** therapeutic agents are being investigated in vitro and in vivo for use in treating human immunodeficiency virus infection, hepatitis B virus infection, herpes simplex virus infection, papillomavirus infection, cancer, restenosis, rheumatoid arthritis, and allergic disorders. Although many results are preliminary, some are promising and have led to clinical trials. A major goal in developing methods of delivering **antisense** agents is to reduce their susceptibility to nucleases while retaining their ability to bind to targeted sites. Modification of the phosphodiester linkages in oligonucleotides can lend the sequences enzymatic stability without affecting their binding capacities. Carrier systems designed to protect the **antisense** structure and improve passage through the cell membrane include liposomes, water-soluble polymers, and nanoparticles. The pharmacokinetics of **antisense** agents are under investigation. **Antisense** therapeutic agents have the potential to become an integral part of medicinal regimens.

L15 ANSWER 29 OF 69 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 1995:938238 CAPLUS

DOCUMENT NUMBER: 123:329975
TITLE: Antiviral transgenic plants, vectors, cells and methods
INVENTOR(S): Silverman, Robert H.; Sengupta, Dibyendu N.
PATENT ASSIGNEE(S): Cleveland Clinic Foundation, USA
SOURCE: PCT Int. Appl., 196 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 4
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|---|---|---|---|---|
| WO 9522245 | A1 | 19950824 | WO 1995-US2058 | 19950216 |
| W: AT, AU, BB, BG, BR, BY, CA, CH, CZ, DE, DK, ES, FI, GB, HU, JP, KP, KR, KZ, LK, LU, MG, MN, MW, NL, NO, NZ, PL, PT, RO, RU, SD, SE, SK, UA, VN | | | | |
| RW: AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG | | | | |
| CA 2183461 | AA | 19950824 | CA 1995-2183461 | 19950216 |
| AU 9519234 | A1 | 19950904 | AU 1995-19234 | 19950216 |
| AU 706185 | B2 | 19990610 | | |
| EP 753992 | A1 | 19970122 | EP 1995-911802 | 19950216 |
| R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LI, LU, MC, NL, PT, SE | | | | |
| BR 9507425 | A | 19970916 | BR 1995-7425 | 19950216 |
| PRIORITY APPLN. INFO.: | | | US 1994-198973 | 19940218 |
| | | | WO 1995-US2058 | 19950216 |

AB Isolated 2-5A-dependent RNases, an interferon-induced enzyme which is activated by 5'-phosphorylated 2',5'-linked oligoadenylates (2-5A) and implicated in both the mol. mechanisms of interferon action and in the fundamental control of RNA stability in mammalian cells, and encoding sequences therefore are disclosed. The expression cloning and anal. of murine and human 2-5A-dependent RNases is also disclosed. In addn., recombinant nucleotide sequences, recombinant vectors, recombinant cells and antiviral plants which express, for example, 2-5A-dependent RNase, 2-5A synthetase and/or **double-stranded RNA** dependent protein kinase (PKR), or other amino acid sequences which have activity that interferes with or inhibits viral replication are disclosed.

L15 ANSWER 30 OF 69 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.
ACCESSION NUMBER: 95280588 EMBASE
DOCUMENT NUMBER: 1995280588
TITLE: Involvement of the **double-stranded-RNA**-dependent kinase PKR in interferon expression and interferon-mediated antiviral activity.
AUTHOR: Der S.D.; Lau A.S.
CORPORATE SOURCE: Div. of Pediatric Infect. Diseases, Department of Pediatrics, San Francisco General Hospital, 1001 Potrero Avenue,San Francisco, CA 94110, United States
SOURCE: Proceedings of the National Academy of Sciences of the United States of America, (1995) 92/19 (8841-8845).
ISSN: 0027-8424 CODEN: PNASA6
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
026 Immunology, Serology and Transplantation
029 Clinical Biochemistry
037 Drug Literature Index
LANGUAGE: English
SUMMARY LANGUAGE: English

AB The signaling mechanisms responsible for the induced expression of interferon (IFN) genes by vital infection or **double-stranded RNA** (**dsRNA**) are not well understood.

Here we investigate the role of the interferon-induced **dsRNA** -dependent protein kinase PKR in the regulation of IFN induction. Biological activities attributed to PKR include regulating protein synthesis, mediating IFN actions, and functioning as a possible tumor suppressor. Since binding of **dsRNA** is required for its activation, PKR has been considered as a candidate signal transducer for regulating IFN expression. To examine this role of PKR, loss-of-function phenotypes in stable transformants of promonocytic U-937 cells were achieved by two different strategies, overexpression of an **antisense** PKR transcript or a dominant negative PKR mutant gene. Both types of PKR-deficient cells were more permissive for viral replication than the control U-937 cells. As the result of PKR loss, they also showed impaired induction of IFN-.alpha. and IFN-.beta. genes in response to several inducers-specifically, encephalomyocarditis virus, lipopolysaccharide, and phorbol 12-myristate 13-acetate. Interestingly, while IFN-.alpha. induction by **dsRNA** was impaired in PKR-deficient cells, IFN-.beta. induction remained intact. Loss of PKR function also resulted in decreased antiviral activity as elicited by IFN-.alpha. and, to a greater extent, by IFN- .gamma.. These results implicate PKR in the regulation of several antiviral activities.

=> s cryptosporidium parvum

L16 4975 CRYPTOSPORIDIUM PARVUM

=> s l16 and antisense

L17 2 L16 AND ANTISENSE

=> d l17 ibib abs tot

L17 ANSWER 1 OF 2 CAPLUS COPYRIGHT 2001 ACS
ACCESSION NUMBER: 2000:790338 CAPLUS
DOCUMENT NUMBER: 133:361903
TITLE: Anti-microbial agents, diagnostic reagents, and vaccines based on unique Apicomplexan parasite components
INVENTOR(S): McLeod, Rima W.; Roberts, Craig; Roberts, Fiona; Johnson, Jennifer; Kirisits, Michael; Ferguson, David; Lyons, Russell; Mui, Ernest; Haselkorn, Robert; Mack, Doug; Samuel, Benjamin; Gornicki, Piotri; Zuther, Ellen
PATENT ASSIGNEE(S): Arch Development Corporation, USA; MRJ Trust
SOURCE: PCT Int. Appl., 251 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|---|---|---|---|---|
| WO 2000066154 | A2 | 20001109 | WO 2000-US11478 | 20000427 |

W: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CR, CU, CZ, DE, DK, DM, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
RW: GH, GM, KE, LS, MW, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG

PRIORITY APPLN. INFO.: US 1999-132506 19990504

AB This invention relates uses of components of plant-like metabolic pathways not including psbA or PPi phosphofructokinase and not generally operative in animals or encoded by the plastid DNA, to develop compns. that interfere with Apicomplexan growth and survival. Components of the pathways include enzymes, transit peptides and nucleotide sequences encoding the enzymes and peptides, or promoters of these nucleotide sequences to which antibodies, **antisense** mols. and other inhibitors are directed. Diagnostic and therapeutic reagents and vaccines are developed based on the components and their inhibitors. A cDNA sequence that encodes chorismate synthase expressed at an early state of Apicomplexan development, is disclosed and may be altered to produce a "knockout" organism useful in vaccine prodn.

L17 ANSWER 2 OF 2 CAPLUS COPYRIGHT 2001 ACS

ACCESSION NUMBER: 1998:89364 CAPLUS
DOCUMENT NUMBER: 128:165312
TITLE: The plant-like structural proteins and metabolic pathways of apicomplexan parasites and the development of diagnostic and therapeutic reagents
INVENTOR(S): McLeod, Rima L. W.; Roberts, Craig W.; Roberts, Fiona; Johnson, Jennifer J.; Mets, Laurens
PATENT ASSIGNEE(S): Arch Development Corp., USA; McLeod, Rima L. W.; Roberts, Craig W.; Roberts, Fiona; Johnson, Jennifer J.; Mets, Laurens
SOURCE: PCT Int. Appl., 212 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|---|---|---|---|---|
| WO 9803661 | A2 | 19980129 | WO 1997-US12497 | 19970718 |
| WO 9803661 | A3 | 19981008 | | |
| W: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GE, HU, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, TJ, TM, TR, TT, UA, UG, US, US, US, US, UZ, VN, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM | | | | |
| RW: GH, KE, LS, MW, SD, SZ, UG, ZW, AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG | | | | |
| AU 9740411 | A1 | 19980210 | AU 1997-40411 | 19970718 |
| EP 918868 | A2 | 19990602 | EP 1997-937983 | 19970718 |
| R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, FI | | | | |

PRIORITY APPLN. INFO.:

| | |
|---|---|
| US 1996-22209 | 19960719 |
| US 1996-773302 | 19961223 |
| US 1997-40849 | 19970317 |
| US 1997-49620 | 19970613 |
| WO 1997-US12497 | 19970718 |

AB Apicomplexan parasites have been found to have a no. of structural proteins and metabolic pathways showing greater similarity to the plant homologs than the animal ones. These proteins and pathways can be used as targets for the diagnosis and treatment of infection with greater specificity for the parasite with lowered risks of complications for the carrier. Suitable targets include enzymes, transit peptides, their genes or promoters. Therapeutic agents include antibodies, **antisense** nucleic acids, and enzyme inhibitors. In vitro inhibitor assays identified a no. of pathways: heme biosynthesis, alternative oxidase,

glyoxylate cycle, and chorismate biosynthesis, thought to be absent from animals. Herbicides active against these pathways were tested and found to inhibit a no. of Apicomplexans. There was some synergism when inhibitors were used in combination. An EST clone from Toxoplasma gondii was found to have sequence similarity to tomato chorismate synthase.

=> s l16 and (dsrna or (double stranded Rna)

UNMATCHED LEFT PARENTHESIS 'AND (DSRNA'
The number of right parentheses in a query must be equal to the number of left parentheses.

=> s l16 and (dsrna or (double stranded Rna))

L18 27 L16 AND (DSRNA OR (DOUBLE STRANDED RNA))

=> dup rem l18

PROCESSING COMPLETED FOR L18
L19 8 DUP REM L18 (19 DUPLICATES REMOVED)

=> d l19 ibib abs tot

L19 ANSWER 1 OF 8 MEDLINE DUPLICATE 1
ACCESSION NUMBER: 2000387814 MEDLINE
DOCUMENT NUMBER: 20304968
TITLE: Association of RNA polymerase complexes of the parasitic protozoan **Cryptosporidium parvum** with virus-like particles: heterogeneous system.
AUTHOR: Khramtsov N V; Upton S J
CORPORATE SOURCE: Division of Biology, Kansas State University, Manhattan 66506-4901, USA.. podolsk@ksu.edu
CONTRACT NUMBER: 1RO1AI/DK42545-01A1 (NIAID)
SOURCE: JOURNAL OF VIROLOGY, (2000 Jul) 74 (13) 5788-95. Journal code: KCV. ISSN: 0022-538X.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Cancer Journals
ENTRY MONTH: 200010
ENTRY WEEK: 20001002
AB RNA polymerase complexes were purified from **Cryptosporidium parvum**, a parasitic protozoan known to infect many species of mammals including humans. Western blot analysis revealed the association of the complexes with two different proteins, encoded by large and small segments of viral double-stranded RNAs. Each complex was found to contain only **double-stranded RNA**, both double- and single-stranded RNA, or only single-stranded RNA. Maximum RNA-dependent RNA polymerase activity was observed within the complexes containing both double- and single-stranded RNAs. These complexes possessed both transcriptase and replicase polymerase activities. Virus-like particles with a diameter of 31 nm were copurified with RNA polymerase complexes, and buoyant density and polymerase studies suggest that C. parvum harbors a putative **double-stranded RNA** virus which separately encapsidates the large and small RNA segments. The mechanism of replication and other characteristics of this virus are similar to those of the viruses of the family Partitiviridae, previously identified only in fungi and plants.

L19 ANSWER 2 OF 8 MEDLINE DUPLICATE 2
ACCESSION NUMBER: 2000424041 MEDLINE
DOCUMENT NUMBER: 20336413

TITLE: Development of a novel, rapid integrated **Cryptosporidium parvum** detection assay.
AUTHOR: Kozwich D; Johansen K A; Landau K; Roehl C A; Woronoff S; Roehl P A
CORPORATE SOURCE: Xtrana Inc., Denver, Colorado 80230, USA.
SOURCE: APPLIED AND ENVIRONMENTAL MICROBIOLOGY, (2000 Jul) 66 (7) 2711-7.
Journal code: 6K6. ISSN: 0099-2240.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200011
ENTRY WEEK: 20001102

AB The aim of this study was to develop a reverse transcription-PCR assay and lateral flow detection protocol for specific identification of **Cryptosporidium parvum**. The method which we developed is sensitive and specific and has a low limit of detection. In our protocol a solid phase material, the Xtra Bind Capture System, was used for extraction and purification of **double-stranded RNA** (**dsRNA**) specific for C. parvum. The Xtra Bind Capture System interfaced with pellets concentrated from water samples collected with previously developed filtration devices. The pellets were resuspended in reagent water (final volume, 0.5 ml), and an equal amount of rupture buffer and the Xtra Bind Capture System was added to the resuspended pellet mixture. The **dsRNA** target sequences in a 0.5-ml portion were captured by the solid phase material via hybridization. The debris and potential inhibitors were removed by washing the Xtra Bind material several times with buffer. The Xtra Bind material with its bound **dsRNA** was added directly to an amplification reaction mixture, and the target was amplified without elution from the Xtra Bind material. A PCR was performed in the presence of the Xtra Bind Capture System, which resulted in robust amplification of the target. The detection system which we used was adapted from lateral flow chromatography methods typically used for antigen-antibody reactions. The result was a colored line that was visible if the organism was present. When this method was used, we were able to reproducibly and correctly identify 10 oocysts added to 0.5 ml of reagent water. When the protocol was evaluated with a small set of environmental samples, the level of detection was as low as 1 oocyst/liter. The total time from resuspension of the pellet to detection was about 3 h, which is considerably less than the 5 h required for immunomagnetic separation followed by an indirect immunofluorescence assay and microscopy.

L19 ANSWER 3 OF 8 MEDLINE DUPLICATE 3
ACCESSION NUMBER: 2000241457 MEDLINE
DOCUMENT NUMBER: 20241457
TITLE: Presence of double-stranded RNAs in human and calf isolates of **Cryptosporidium parvum**.
AUTHOR: Khramtsov N V; Chung P A; Dykstra C C; Griffiths J K; Morgan U M; Arrowood M J; Upton S J
CORPORATE SOURCE: Division of Biology, Kansas State University, Manhattan 66506, USA.
CONTRACT NUMBER: 1RO1AI/DK42545-01A1 (NIAID)
R825148-01-0
SOURCE: JOURNAL OF PARASITOLOGY, (2000 Apr) 86 (2) 275-82.
Journal code: JL3. ISSN: 0022-3395.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals

OTHER SOURCE: GENBANK-U11761
ENTRY MONTH: 200006
ENTRY WEEK: 20000605
AB We examined the occurrence of 2 virus-like double-stranded (ds)RNAs in human and calf isolates of **Cryptosporidium parvum** senso latu and other microorganisms, including 7 other members of the genus. A total of 32 isolates of C. parium, 16 from humans (5 from acquired immune deficiency syndrome patients) and 16 from calves, were analyzed. Ethidium bromide staining, or Northern blot analysis, or reverse transcription/polymerase chain reaction, or all 3 methods, revealed that both genotype 1 and genotype 2 isolates of C. parvum possessed these dsRNAs. No other Cryptosporidium spp. or other organisms examined possessed these dsRNAs. Comparison analysis of partial cDNA sequences of dsRNAs from human and calf isolates revealed a high degree of similarity (>92% and >93% identical nucleotides for large and small dsRNAs, respectively). Slight, consistent differences in nucleotide sequences could be seen at select sites and were associated with an isolate being either genotype 1 or 2. Because of the widespread distribution of the dsRNAs, the similarity of these molecules between isolates, and high host specificity, these nucleic acids may prove to represent species-specific molecular markers for C. parvum. Evidence also suggests that the **dsRNA** can be utilized for molecular genotyping of C. parvum.

L19 ANSWER 4 OF 8 BIOSIS COPYRIGHT 2001 BIOSIS
ACCESSION NUMBER: 2001:8235 BIOSIS
DOCUMENT NUMBER: PREV200100008235
TITLE: Tracking **Cryptosporidium parvum** parasites by sequence analysis of the small **double -stranded RNA.**
AUTHOR(S): Xiao, L. (1); Limor, J. R. (1); Bern, C. (1); Lal, A. A. (1)
CORPORATE SOURCE: (1) Division of Parasitic Diseases, National Center for Infectious Diseases, Centers for Disease Control and Prevention, Atlanta, GA USA
SOURCE: American Journal of Tropical Medicine and Hygiene, (March, 2000) Vol. 62, No. 3 Supplement, pp. 261. print.
Meeting Info.: 49th Annual Meeting of the American Society of Tropical Medicine and Hygiene Houston, Texas, USA October 29-November 02, 2000 American Society of Tropical Medicine and Hygiene
. ISSN: 0002-9637.
DOCUMENT TYPE: Conference
LANGUAGE: English
SUMMARY LANGUAGE: English

L19 ANSWER 5 OF 8 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.
ACCESSION NUMBER: 2000092093 EMBASE
TITLE: Preliminary profile of the **Cryptosporidium parvum** genome: An expressed sequence tag and genome survey sequence analysis.
AUTHOR: Strong W.B.; Nelson R.G.
CORPORATE SOURCE: R.G. Nelson, Division of Infectious Diseases, San Francisco
General Hospital, San Francisco, CA, United States.
malaria@itsa.ucsf.edu
SOURCE: Molecular and Biochemical Parasitology, (15 Mar 2000) 107/1
(1-32).
Refs: 103
ISSN: 0166-6851 CODEN: MBIPDP
PUBLISHER IDENT.: S 0166-6851(99)00225-X
COUNTRY: Netherlands
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
LANGUAGE: English

SUMMARY LANGUAGE: English

AB **Cryptosporidium parvum** is a protozoan enteropathogen that infects humans and animals and causes a pronounced diarrheal disease that can be life- threatening in immunocompromised hosts. No specific chemo- or immunotherapies exist to treat cryptosporidiosis and little molecular information is available to guide development of such therapies.

To accelerate gene discovery and identify genes encoding potential drug and vaccine targets we constructed sporozoite cDNA and genomic DNA sequencing libraries from the Iowa isolate of C. parvum and determined .apprx.2000 sequence tags by single-pass sequencing of random clones. Together, the 567 expressed sequence tags (ESTs) and 1507 genome survey sequences (GSSs) totaled one megabase (1 mb) of unique genomic sequence indicating that .apprx.10% of the 10.4 mb C. parvum genome has been sequence tagged in this gene discovery expedition. The tags were used to search the public nucleic acid and protein databases via BLAST analyses, and 180 ESTs (32%) and 277 GSSs (18%) exhibited similarity with database sequences at smallest sum probabilities P(N).ltoreq.10-8. Some tags encoded proteins with clear therapeutic potential including S-adenosylhomocysteine hydrolase, histone deacetylase, polyketide/fatty-acid synthases, various cyclophilins, thrombospondin-related cysteine-rich protein and ATP-binding- cassette transporters. Several anonymous ESTs encoded proteins predicted to contain signal peptides or multiple transmembrane spanning segments suggesting they were destined for membrane-bound compartments, the cell surface or extracellular secretion. One-hundred four simple sequence repeats were identified within the nonredundant sequence tag collection with (TAA)(.gtoreq.6)/(TTA)(.gtoreq.6) and (TA)(.gtoreq.10)/(AT)(.gtoreq.10) being the most prevalent, occurring 40 and 15 times, respectively. Various cellular RNAs and their genes were also identified including the small and large ribosomal RNAs, five tRNAs, the U2 small nuclear RNA, and the small and large virus-like, double- stranded RNAs. This investigation has demonstrated that survey sequencing is an efficient procedure for gene discovery and genome characterization and has identified and sequence tagged many C. parvum genes encoding potential therapeutic targets. (C) 2000 Elsevier Science B.V.

L19 ANSWER 6 OF 8 MEDLINE DUPLICATE 4
ACCESSION NUMBER: 2000129497 MEDLINE
DOCUMENT NUMBER: 20129497
TITLE: Genomic characterisation of the large segment of a rabbit picobirnavirus and comparison with the atypical picobirnavirus of **Cryptosporidium parvum**.
AUTHOR: Green J; Gallimore C I; Clewley J P; Brown D W
CORPORATE SOURCE: Enteric and Respiratory Virus Laboratory, Central Public Health Laboratory, London, UK.
SOURCE: ARCHIVES OF VIROLOGY, (1999) 144 (12) 2457-65. Journal code: 8L7. ISSN: 0304-8608.
PUB. COUNTRY: Austria
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Cancer Journals
OTHER SOURCE: GENBANK-AJ244022
ENTRY MONTH: 200004
ENTRY WEEK: 20000404

AB The 2362 base pair sequence of the larger of the two **double stranded RNA** genome segments of a rabbit strain of picobirnavirus (PBV) has a major open reading frame (ORF) of 591 amino acids and two smaller ORFs of 55 and 155 amino acids. A clone of the segment did not hybridise with other viral bisegmented ds RNAs from faecal samples. There is no relationship in sequence or organisation between this

PBV sequence and the bisegmented dsRNAs found associated with **Cryptosporidium parvum**. This suggests that there are at least two distinct classes of bisegmented **dsRNA** viruses or viral-like agents in faeces.

L19 ANSWER 7 OF 8 CAPLUS COPYRIGHT 2001 ACS DUPLICATE 5
ACCESSION NUMBER: 1998:390391 CAPLUS
DOCUMENT NUMBER: 129:132082
TITLE: High-temperature inducible cell-free transcription and replication of double-stranded RNAs within the parasitic protozoan **Cryptosporidium parvum**
AUTHOR(S): Khramtsov, Nikolai V.; Upton, Steve J.
CORPORATE SOURCE: Division of Biology, Kansas State University, Manhattan, KS, 66506, USA
SOURCE: Virology (1998), 245(2), 331-337
CODEN: VIRLAX; ISSN: 0042-6822
PUBLISHER: Academic Press
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Sporozoites of the protozoan parasite, **Cryptosporidium parvum**, were found to contain free, full-size plus strands transcribed from two extrachromosomal, cytoplasmic, virus-like double-stranded RNAs (dsRNAs). Cell-free transcription and replication of both dsRNAs were obsd. in crude sporozoite lysates. RNA polymerase activity was dependent upon addn. of Mg2+ or Mn2+, as well as the four ribonucleoside triphosphates, and was insensitive to inhibitors of cellular DNA-dependent RNA polymerase. Semiconservative transcription of the dsRNAs (plus strand synthesis) was obsd. at a wide range of temps., with an optimum of 50.degree.. In contrast, replication (minus strand synthesis) was detected only at 50 and 60.degree.. (c) 1998 Academic Press.

L19 ANSWER 8 OF 8 MEDLINE DUPLICATE 6
ACCESSION NUMBER: 1998043502 MEDLINE
DOCUMENT NUMBER: 98043502
TITLE: Virus-like, double-stranded RNAs in the parasitic protozoan **Cryptosporidium parvum**.
AUTHOR: Khramtsov N V; Woods K M; Nesterenko M V; Dykstra C C; Upton S J
CORPORATE SOURCE: Division of Biology, Kansas State University, Manhattan 66506, USA.. podolsk@ksu.edu
SOURCE: MOLECULAR MICROBIOLOGY, (1997 Oct) 26 (2) 289-300.
Journal code: MOM. ISSN: 0950-382X.
PUB. COUNTRY: ENGLAND: United Kingdom
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-U95995; GENBANK-U95996
ENTRY MONTH: 199804
AB We have discovered and analysed two novel, linear extrachromosomal double-stranded RNAs (dsRNAs) within oocysts of major north Amercian isolates of **Cryptosporidium parvum**, a parasitic protozoan that infects the gastrointestinal tract of a variety of mammals, including humans. These dsRNAs were found to reside within the cytoplasm of sporozoites, and were not detected in other species of the genus. cDNAs representing both **dsRNA** genomes were cloned and sequenced, 1786 and 1374 nt, and each encoded one large open reading frame (ORF). The deduced protein sequence of the larger **dsRNA** (L-**dsRNA**) had homology with viral RNA-dependent RNA polymerases (RDRP), with more similarity to polymerases from fungi than those from other protozoa. The

deduced protein sequence from the smaller **dsRNA** (S-**dsRNA** ) had limited similarity with mitogen-activated c-June NH2 terminal protein kinases (JNK) from mammalian cells. Attempts to visually identify or purify virus-like particles associated with the dsRNAs were unsuccessful. Sensitivity of the dsRNAs to RNase A also suggests that the dsRNAs may be unencapsidated. A RDRP activity was identified in crude extracts from C. parvum sporozoites and products of RNA polymerase activity derived in vitro were similar to the dsRNAs purified directly from the parasites.